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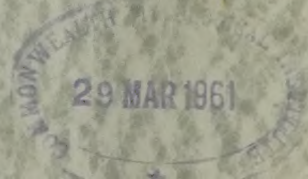
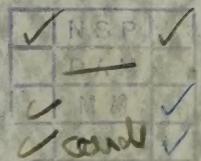
長尾研究所菌類研究報告 7

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概 観 2 0 年

長尾研究所は、商工省から財団法人として 1941 年 1 月 24 日認可された研究所である。財団の理事には長尾欽弥氏の他、斎藤賢道、薮田貞治郎、柴田桂太、真島利行、島峯 徹、青木 保、坂口康蔵、佐々木喬、慶松勝左衛門、志賀 潔、岡部連の諸博士が、又監事には関屋貞三郎、石井光雄の両氏がそれぞれ就任し、特に斎藤、薮田両博士は、研究所開設準備に当つて特に貢献するところがあつた。

1942 年、研究項目「エレモテシウム・アッシュビー菌によるフラビンの生産」の成果が学界に発表せられた。

1943 年、前東京帝国大学農学部教授小南清氏が主任研究員として、又大阪帝国大学教授理学博士赤堀四郎及び京都帝国大学教授工学博士高田亮平の両氏が参与としてそれぞれ本所の研究陣に参加した。

1944 年 2 月 5 日天皇陛下には宮城内生物学御研究所に於て、服部広太郎博士の御説明に依り、フラビン生産菌エレモテシウム・アッシュビー並びに、該菌を応用して製造したビタミン B₂ 剤「ワカフラビン」を御覧になり、これらを受納せられた。

同年同月、軍に依り、陸軍軍医学校内にペニシリン委員会が組織せられ、本研究所からは主任研究員斎藤賢道、同 薮田貞治郎、同 小南 清、研究員根平武雄の四氏が委員に任命され、本研究所は、直ちに同委員会に青黴属合計 420 菌株を提出した。同委員会には全国から二千余種の菌株が集つたが、ペニシリン生産菌は三株で、その二株が本研究所の提出にかかるものであつた。

1945 年 4 月から終戦に至るまでの間、本研究所は陸軍軍医学校の要請に依り前後四回に亘つてペニシリン講習会を開催した。

1946 年 1 月、戦後の新状況に応え、京都支所（斎藤賢道博士研究室）、滝野川支所（柴田桂太博士研究室）、伊丹支所（赤堀四郎博士研究室）、甲陽支所（船久保英一博士研究室）及び東京長尾研究所内に附属診療所（長谷川和三博士主任）が開設されそれぞれ業務を開始した。また同年月には長尾研究所編図説科学叢書「ペニシリン絵物語」及び「うまい菌と毒菌」の二冊が刊行された。然しこれら新企画はいずれも戦後経済状況変動の余波を受け、半途にしてその継続を断念せざるを得なくなつた。

1949 年本研究所はアスカリドールの駆虫効力を保存してその副作用を抑制する研究を完成した。

1950 年本研究所は文部省並びに International Association of Microbiologist の推薦に基き、9 月 8 日附を以て International Federation of Culture Collections of Micro-Organisms に正式会員として加盟した。これを機に日本国内に於ても菌株保存連盟が結成せられ、本所理事・主任研究員小南清氏が初代会長に就任した。この前後から菌株の交換も活発に行われるようになってきた。

従来本所は通産省の所轄下にあつたが 1951 年度より通産・文部両省の共管下に立つこととなるに伴い、本財団寄附行為を新時代の理念に副うよう改訂し、理事長・所長及び専務理事の制を新たに定め、初代理事長には財団寄附者長尾欽弥氏が、同じく所長には理事小南清氏がそれぞれ就任した。

1952 年 5 月には多年の宿願であつた機関誌 NAGAOA 第 1 号が発刊せられた。

1954 年、稲芯枯線虫防除薬に関する研究が完成した。

本研究所は戦時中より大豆を原料とする高蛋白食品の研究を手がけ来つたが、その後さらに研究・改良を加えたものに就ては 1959 年特許権を得た。

1958 年 11 月 28 日、本研究所非常勤研究員福島博氏は第三次南極地域観測隊に参加した。

1959 年 10 月、本研究所は由緑の地、東京都品川区北品川より、新たに世田ヶ谷区三宿町 380 番地に移転した。爾後施設整備を鋭意急ぎつつも、基礎部門・応用部門の研究は一日もこれを忽がせにすることなく、所員一同その業務に専念している。

(1960. 12. 20)

Jafnea, a New Genus of the *Pezizaceae*

By Richard P. KORF*

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In 1949 I had the pleasure of several long discussions on discomycete problems with Professor J.A.F. Nannfeldt in his laboratory at Uppsala University. In discussing the genus *Helvella* Fr. and its allies, which make up the tribe Acetabuleae (Nannfeldt, 1937, 1938), we were in agreement that the two North American species treated by Durand (1906) in *Macropodia* Fuckel and by Seaver (1928) in *Paxina* Kuntze have nothing to do with the other species placed there (mostly species of *Helvella* to our mind). He suggested to me that these two species might well deserve a genus of their own, but that since he had not seen them in living condition, it would be more appropriate for an American to work on the problem.

Five years later I proposed the generic name *Jafnea* in his honor for these species (Korf, 1954), but no diagnosis was published and the name remained a *nomen nudum*. At that time I had seen only the type species in living condition. Only once in the intervening six years have I seen living material of the second American species. It is apparently common in the mid-western United States but very rare in our north-eastern flora. In addition I have now collected four times an apparently undescribed species from Japan which I place in the genus.

The new genus is characterized by the possession of a rather thick and most characteristic excipulum. The ectal excipulum consists of cells arranged in parallel rows, each cell elongated perpendicularly to the outer surface of the apothecium. The outermost cells are brown, while the inner cells of the ectal layer may be brown (Fig. 1, 2) or nearly hyaline (Fig. 3). I know of but one genus of the *Pezizaceae* in which a somewhat similar tissue structure occurs, *Sowerbyella* Nannf., but that genus appears to be quite distant from *Jafnea*. It is something of a surprise to me to find that the genus which appears to be most closely related is *Genea* Vitt., an unquestioned member of the *Tuberales*! (Compare Fischer, 1897 : Fig. 204 A, 1938 : Fig. 6 A with Fig. 2, 3 of this paper.)

* The research reported here was primarily carried out under National Science Foundation Grant G 9041, "Discomycete Flora of Asia." Japanese collections were in part made while the writer was Fulbright Research Professor and National Science Foundation Senior Postdoctoral Fellow at Yokohama National University, 1957-58. Some American collections were aided by Cornell University Faculty Research Grant 5276. The writer is indebted to Mr. James Kimbrough for technical assistance, Mr. Howard Lyon for photography, and Dr. William Dress for preparation of the Latin diagnoses.

At the base of the apothecia are branching, woolly hyphae (Fig. 1) which enmesh particles of soil and debris to form a columnar mass or pseudostipe. It is presumably this character which led Durand (1906) and Seaver (1928) to include the American species in the helvelloid series. The structure of the excipulum is so widely divergent from other species of "*Macropodia*" and "*Paxina*" (i.e., cupulate *Helvella*) that the species must surely belong not only to another genus but to another tribe. (The ascospores of *Jafnea* are also equally ill-fitting for the Acetabuleae.)

Just where *Jafnea* will fit best within the Pezizaceae remains problematical. The two American species are provided with short, brown, superficial, excipular outgrowths which might well be termed hairs (Fig. 2, 3), though these are lacking in the new Japanese species. The nearly hyaline basal hyphae in all three species are very reminiscent of similar flexuous hairs in the genus *Sepultaria* (Cooke) Boud. and in some species of *Trichophaea* Boud. Saccardo (1889), in fact, included the two known species in *Lachnea* Fr. non L. on the basis of the pilose excipulum. Despite the fact that *Jafnea* does not seem closely related to either *Humaria* Fuckel *sensu stricto** or to *Sepultaria*, I am inclined to include *Jafnea* for the time being in the admittedly heterogeneous tribe Ciliarieae (Korf, 1954, 1958 : I).

The ascospores of the new genus are also characteristic, being large, and marked at maturity with warts (Fig. 4, 5, 6) which stain deeply in cotton-blue dyes when heated (Korf, 1952, 1958 : I, 1960). In youth the spores have two large guttules (Fig. 7, 8, 9), but these disappear at maturity. In the new Japanese species (Fig. 4) the markings can be seen to be limited by a membrane enclosing also an only slightly staining exospore material similar to that previously described in other Pezizaceae by Le Gal (1947). The warts tend to be largest near the ends of the spore. This is particularly evident in the type species, where they take the form of cushion-shaped apiculi (Fig. 6).

The genus is known thus far only from North America, Japan, and India (Batra, 1960). The following key points out the major distinguishing characters among the species.

- A. Apothecia deep-cupulate; hymenium and excipulum some shade of tan; exposed excipulum covered with short, brown hairs.
 - 1. Apothecia small, usually not exceeding 2 cm. diam.; ascospores fusoid, lightly marked, often apiculate, (29-) 30.8-37.1 (-44) \times 9.1-10.9(-11.8) μ . (North America, Japan, India) **J. fusicarpa**
 - 2. Apothecia large, usually 2-5 cm. diam.; ascospores fusiform-ellipsoid, more

* I join with Denison (1956, 1960) in designating *Humaria hemisphaerica* (Wigg. ex Gray) Fekl. as the **LECTOTYPE** of *Humaria* Fekl., Jahrb. Nass. Ver. Naturk. 23-24 : 320. 1870.

strongly marked, non-apiculate, $23.5-30.8(-33) \times 9.1-12.7 \mu$. (North America)

.....**J. semitosta**

- B. Apothecia cupulate or turbinate when young, discoid at maturity; hymenium light brown to brownish green, excipulum reddish brown to drab greenish brown; exposed excipulum devoid of hairs, distinctly pustulate; apothecia small, not exceeding 1.5 cm. diam.; ascospores ellipsoid, with huge warts, (19-) $20.8-23.5 (-25.3) \times (9.1-) 10-11.8 (-12.7) \mu$. (Japan).....**J. imaii**

JAFNEA Korf, gen. nov. (Pezizaceae, Ciliarieae)

Apothecia discoid to deep cupulate, 0.5-8 cm. diam.; ectal excipulum composed of cells elongated perpendicularly to the outer surface of the apothecium, outermost cells brown-walled, inner cells hyaline or brown; hairs absent, or if present superficial, short, brown; basal hyphae abundant, nearly hyaline, enmeshing particles of soil to form a pseudostipe or cushion; asci operculate, 8-spored, J-; ascospores large, hyaline, biguttulate in youth, non-guttulate at maturity, marked with small to large warts. On soil and duff.

ETYMOLOGY: A cryptogram from the initials of Professor J.A.F. Nannfeldt, +*-ea* for euphony. Gender: feminine.

SPECIES HOLOTYPE: *Peziza fusicarpa* Gerard.

Jafnea fusicarpa (Gerard) Korf, comb. nov.

Basionym: *Peziza fusicarpa* Ger., Bull. Torrey Bot. Club 4: 64. 1873.

NOTES: The descriptions by Durand (1906) and Seaver (1928) are adequate, except that neither author described the spore-markings in detail (cfr. Fig. 6). I have examined an authentic specimen of *Peziza fusicarpa* Ger. and type specimens of *P. pubida* Berk. and Curt. in Berk., and *P. morgani* Massee in Morgan in the Durand herbarium at Cornell, and agree with the synonymy presented by Durand. (Seaver also lists *Sepultaria aspera* Clements in synonymy, but I have not seen a type specimen.) The only known Japanese specimen, indistinguishable from North American material, was sent to me by Dr. K. Tubaki: HONSHU, Yamagata: Fungi of Japan 1500, CUP, TNS, R.P.K. (Tubaki 45-25).

Jafnea semitosta (Berk. & Curt. in Berk.) Korf, comb. nov.

Basionym: *Peziza semitosta* Berk. & Curt. in Berk., Grevillea 3: 153. 1875.

NOTES: Again I only wish to add an illustration of the spore markings (Fig. 5) to the descriptions by Durand (1906) and Seaver (1928). I have examined the type specimens of *Peziza semitosta* Berk. & Curt. in Berk. and *P. hainesii* Ellis in the Durand herbarium, and agree with Durand's synonymy. (Seaver lists *Sepultaria*

gigantea Clements as a synonym, but I have seen no type.) The species was abundantly collected by the author and others on the Mycological Society of America foray in Bloomington, Indiana, in 1958, but most of the material is immature. The species is not known from Japan.

***Jafnea imaii* Korf, sp. nov.**

Apothecia turbinate to cupulate when young (3–8 mm. diam.), at maturity discoid, 1–1.5 cm. diam.; *receptacle* light reddish brown to drab greenish brown, pustulate above, below giving rise to hyphae which enmesh soil particles; *disc* light brown with an olive-green tint to light olive-green to brownish green, concave to nearly plane. *In section*: *hymenium* ca. 375 μ thick; *subhymenium* ca. 70 μ thick, of compact, light-brown *textura intricata*, hyphae 3.6–9.1 μ diam.; *medullary excipulum* 0–375 μ thick, of loose, light-brown *textura intricata*, hyphae very thin-walled, 5.4–8.2 μ diam.; *ectal excipulum* 100–190 μ thick, of *textura prismatica* to *textura angularis*, cells brown-walled, mostly 21.7–29 \times 12.7–25.3 μ ; *conical pustules* formed of thicker-walled and more spherical excipular cells, pustules ca. 100–125 μ wide at the base, 40–65 μ high; *basal enmeshing hyphae* branched, nearly hyaline, smooth, 4.5–5.4 μ diam. *Asci* 281–366 \times 12.9–19.3 (–21.4) μ , arising from croziers. *Ascospores* ellipsoid, (19–) 20.8–23.5 (–25.3) \times (9.1–) 10–11.8 (–12.7) μ excluding markings, with large warts (Fig. 4) at maturity, in youth with two large guttules (Fig. 7) which disappear at maturity. *Paraphyses* filiform-clavate, hyaline or slightly colored, about as long as the asci, ca. 3.6 μ diam., septa infrequent. On soil and duff.

ETYMOLOGY: In honor of Professor Sanshi Imai, Yokohama National University, in whose company I obtained all four of the collections I have seen in living condition.

TYPE SPECIMEN: S. Imai, M. Hamada, T. Hongo & R.P. Korf. 23. X. 1957. On duff. Between Kiyomizu Temple and Maruyama Park, Kyoto, Kyoto Pref., HONSHU. Fungi of Japan 184, CUP (HOLOTYPE), TNS, R.P.K. (ISOTYPES).

OTHER SPECIMENS EXAMINED: HONSHU, Chiba: F. of J. 1035, CUP, TNS, R.P.K.; Gumma: F. of J. 1499 (in formalin), TNS, R.P.K.; Shiga: F. of J. 221, CUP, TNS, R.P.K.; Tochigi: F. of J. 61, CUP, TNS, R.P.K.; Yamagata: D. Shimizu, Yonezawa, ca. 350 m., 4. VII. 1958, Korf & Shimizu Disc. Jap. 5 (with illustration), R.P.K.

Latin Diagnoses

***Jafnea* Korf, gen. nov. (Pezizaceae, Ciliarieae)**

Apothecia patellata vel urceolata, 0.5–5(–8) cm. diam., excipulo exteriori e *textura prismatica* vel *angulari*, cellulis perpendiculariter ad apothecii faciem exteriori elongatis, exterioribus brunneis, interioribus hyalinis vel brunneis; pilis superficialibus,

brevibus, brunneis, vel in specie una absentibus; hyphae basales abundantes, subhyalinae, humi granula amplexantes et pseudostipitem isto modo efficientes; asci operculati, octospori, apicibus in Iodo non caerulescentibus; ascosporae grandae, hyalinae, primum biguttulatae deinde non guttulae, verrucis minutis vel grandis ornatae. In humo et residuis vegetabilium. Holotypus : *Peziza fusicarpa* Gerard.

Jafnea imaii Korf, sp. nov.

Apothecia primum cupulata vel turbinata, deinde patellata, 1–1.5 cm. diam., excipulo pallide rubido-brunneo vel sordide viridi-brunneo, supra pustulato, infra humi granula amplexantes hyphas efferente, hymenio pallide brunneo vel brunneo-viride; subhymenium ca. 70 μ latum, e textura intricata pallide brunnea, hyphis 3.6–9.1 μ diam.; excipulum medullare 0–375 μ latum, e textura intricata pallide brunnea, hyphis 5.4–8.2 μ diam.; excipulum exterius 100–190 μ latum, cellulis elongatis parietibus brunneis instructis, plerumque 21.7–29 \times 12.7–25.3 μ ; hyphae basales ramosae, subhyalinae, 4.5–5.4 μ diam.; asci 281–366 \times 12.9–19.3 (–21.4) μ , e lituis nati; ascosporae ellipsoideae, (19–) 20.8–23.5 (–25.3) \times (9.1–) 10–11.8 (–12.7) μ verrucis exclusis, juvenilibus duos guttulos magnos postea evanescentes contentibus, maturis verrucis grandis ornatis (Fig. 4); paraphyses hyalinae vel leviter coloratae, filiformi-clavatae, ca. 3.6 μ diam., septis infrequentibus. In humo et residuis vegetabilium. Holotypus : CUP, Fungi of Japan 184.

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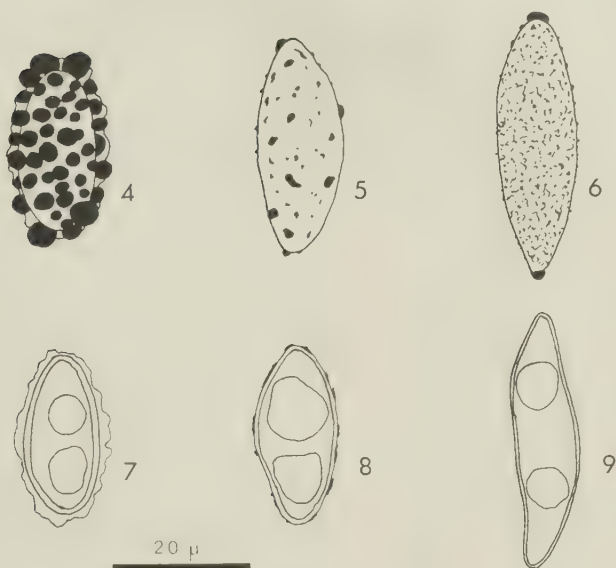
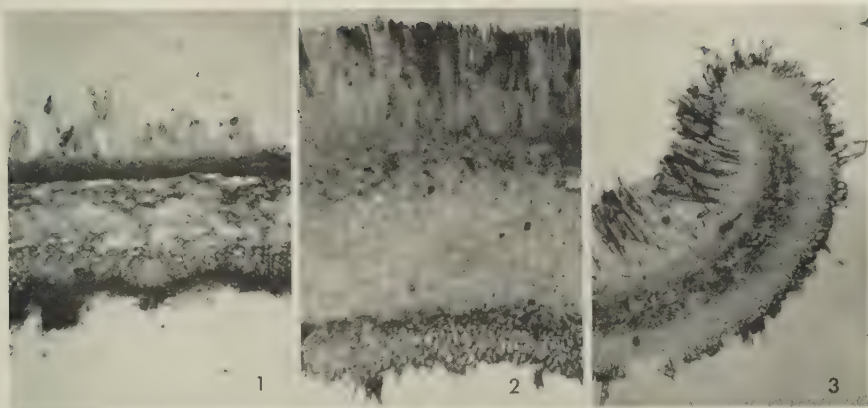
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Pl.1. Korf: *Jafnea*

- Fig. 1-3. Photomicrographs of freezing microtome sections through apothecia of species of *Jafnea*, $\times 50$. Fig. 1, *J. imaii*, showing basal hyphae. Fig. 2, *J. semitosta*. Fig. 3, *J. fusicarpa*.
- Fig. 4-9. Ascospores of species of *Jafnea*, drawn at $\times 1580$ with the aid of a camera lucida and reduced in reproduction to $\times 1000$. Fig. 4-6. Mature ascospores, in surface view, markings deep blue from staining in heated cotton-blue in lactophenol. Fig. 7-9. Immature spores in optical section, before deeply staining markings have appeared. Fig. 4, 7, *J. imaii*. Fig. 5, 8, *J. semitosta*. Fig. 6, 9, *J. fusicarpa*.

Richard P. KORF



On a new yeast genus *Wickerhamia*

by Masami SONEDA

曽根田正己：酵母の一新属ウイッケルハミアについて

In a previous paper (1959), the present author described a new yeast, *Kloeckera fluorescens*, which was isolated from squirrel dung (?) found in the foot of Mt. Fuji. Recently by the suggestion of Dr. L.J. Wickerham, it was found that this yeast produces asci with peculiar ascospores on some agar media. Consequently the present author transfers this to ascosporogenous yeast, establishing a new genus.

Wickerhamia Soneda gen. nov. (Saccharomycetaceae).

Cellulae ovoideae, elongato-ovoidae aut citrifformes. In propagatione vegetativa gemmae polariter formantur. Pseudomycelium paucum. Ascosporae pileiformes lateraliter cum labro, 1, 3, 4, 5, 8, 16 inasco, plerumque 1. Sedimentum, annulus et interdum post aliquot menses insulae formantur. Fermentatio +. Nitras kalicus non assimilatur.

Cells are oval, long oval or lemon shaped. Vegetative reproduction by polar budding on broad base. The cells are liberated from each other in mode of fission. Pseudomycelium primitively developed. The ascospores are cap-shaped; the crown deflects to one side of the sinuous brim, giving the appearance of a sporting cap. The number of ascospores in one ascus is variable, usually one per ascus, often ranging from one up to sixteen (1, 3, 4, 5, 8, 16). In liquid media a sediment and ring are formed. Sometimes islet is formed after a long period. Fermentative. Nitrate is not assimilated. Monotypic genus. Type: *Kloeckera fluorescens* Soneda.

Wickerhamia fluorescens (Soneda) Soneda nov. comb.

Syn. *Kloeckera fluorescens* Soneda, in Nagaoa 6: 16(1959).

Proprieta additamenta. Cultura in agarico pultis avenae (post aliquot dies, 25°C), ascos formantur. Asci elongato-ovoides vel citrifformes, ca $24 \times 28 \mu$ in diam., cum 1-16 (plerumque 1) ascosporis. Ascosporae pileiformes lateraliter cum labro, $3.2 \times 6.4 \mu$ in diam.

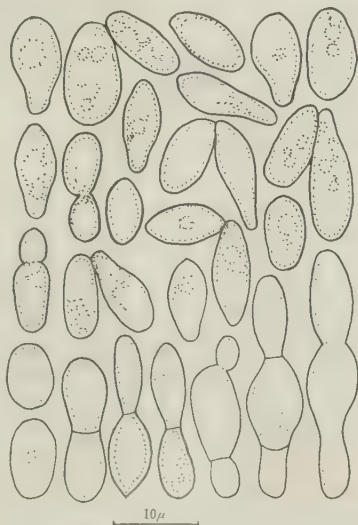


Fig. 1 Vegetative cells of *W. fluorescens* after 3 days in malt extract.

Morphological properties : after 3 days in malt extract at 25°C, cells are long lemon shaped, ovoid or long ovoid $7.2-16 \times 4.2-7.2 \mu$, single or in pairs (Fig. 1), bipolar budding on broad base; sediment and thin ring are formed; after one month at 20°C,

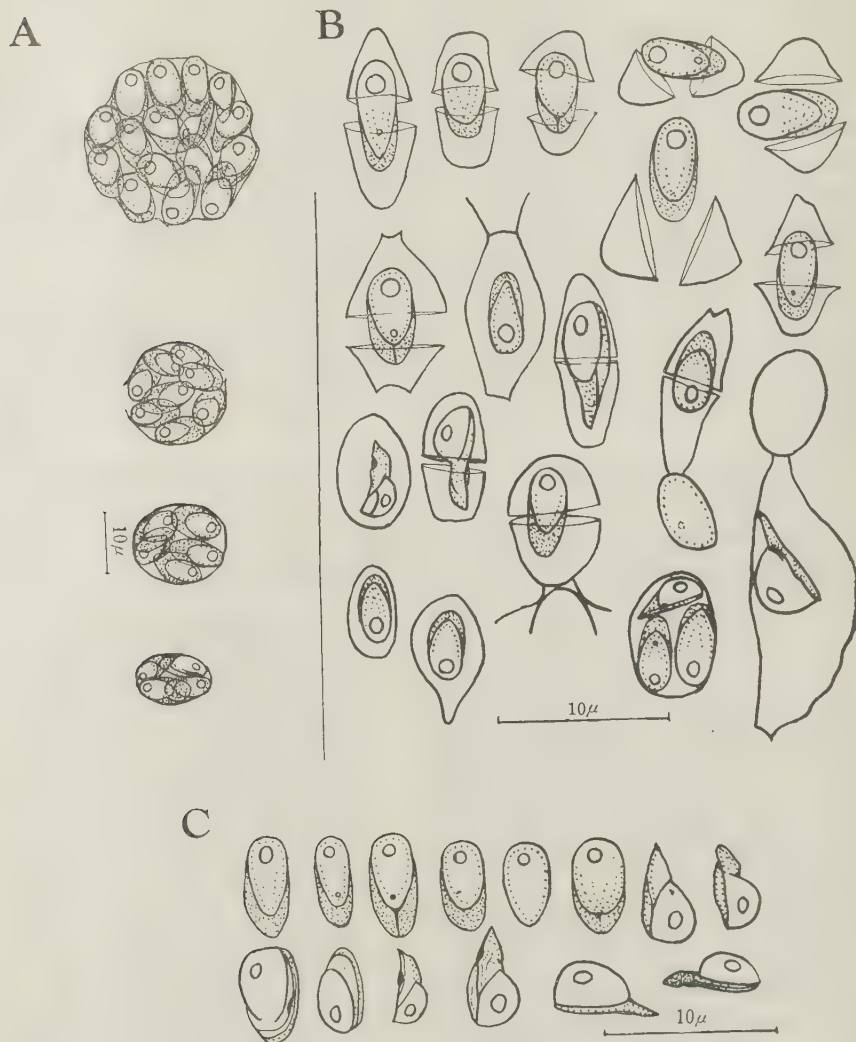


Fig. 2 Asci and ascospores of *W. fluorescens*

- A. Multispored asci; 16, 8, 5 and 4 ascospores respectively in an ascus.
- B. Various types of asci: 1~3 ascospores in an ascus.
- C. Ascospores.

sediment and ring are present, but sometimes some islets are formed, pellicle is not produced. After one month on malt agar at 25°C, the color of streak culture is yellowish to cream, usually strong yellowish because of the diffusion of yellowish pigment in the medium; the streak culture is soft, rather flat, glistening and more or less smooth. Ascus formation is distinct within several days on oat-meal agar* at 25°C.

Ascospores are peculiar cap-shaped, $3.2 \times 6.4 \mu$, crown deflecting to one side of the brim and containing one oil drop. The brim thickened slightly, sinuous and somewhat granulated. The ascospores liberate after sudden rupture of ascus wall (Fig. 2). The ripe asci are long ovoid, elongate ovoid or lemon-shaped, $24 \times 28 \mu$ or more, multispored, containing one to sixteen ascospores, sometimes produce a daughter cell at one pole.

Physiological properties : glucose, galactose and saccharose are fermented, and assimilated, but maltose and lactose are not fermented and assimilated; potassium nitrate is not assimilated; test of ethanol as sole source of carbon is weakly positive; arbutin is not splitted. Type culture is deposited at Nagao Institute (Japan) and Centraal-bureau voor Schimmelcultures, Baarn (Holland).

In 1959, the present author isolated this yeast from the dung of *Calloseiurus erythraeus taiwanensis* (?) which was collected in the foot of Mt. Fuji and established a new species, *Kloeckera fluorescens*. Recently, it appeared that the above yeast reproduces sexually and thus should be transferred from *Kloeckera* to anascosporogenous group. As this species has mechanism of vegetative reproduction by budding on the broad base, this is of the Nadsoniae. The present author gave it a new generic name *Wickerhamia* in memory of Dr. Wickerham who gave him kindly a personal communication about this yeast.

The genus *Wickerhamia* differs from *Nadsonia*, *Saccharomycodes* and *Hanseniaspora* by multi-ascospored ascus and peculiar ascospore. In a process of vegetative reproduction and also in mechanism of sporulation, this is near the above mentioned members of Nadsoniae, making some connective link. The present author speculate that this species may be considered phylogenetically as originated from the *Spermophthoraceae* containing *Spermophthora*, *Ashbya* and *Eremothecium*. Comparison of the ascospore-shape consisting from two parts and the production of riboflavin of *Eremothecium cymbalariae* with those of *Wickerhamia fluorescens* shows a marked similarity between two species. Moreover from the view point of the number of ascospores, *Wickerhamia* may situate at the intermediate position between Nadsoniae and the *Spermophthoraceae*. From the view point of taxonomic significance of the shape of ascospore and of a course of vegetative reproduction, *Wickerhamia* seems not to be

* Oat-meal agar was prepared by extracting 10 g of commercial oat-meal in 100 cc of distilled water at nearly 70°C for 30 min. and kept for 1 hr. at room temperature, then filtered through the gauge and filled up to 200 cc by distilled water to which 2 g of agar was added.

related to the multisporous two genera, *Kluyveromyces* and *Lipomyces*. In addition to the above mentioned properties, *Wickerhamia fluorescens* has strong fermentative ability and can be easily cultivated on common media.

The effect of vitamin on the growth was detected by culturing in a medium of complete set of vitamin except for one. The result is shown in the following table.

Vitamin Requirement of *Wickerhamia fluorescens*

Omitted vitamin (microgram per liter)	Growth
Biotin (2)	—
Calcium pantothenate (400)	+
Inositol (2000)	+
Niacin (400)	+
Para-aminobenzoic acid (200)	+
Pyridoxin hydrochloride (400)	+
Riboflavin (200)	+
Thiamin hydrochloride (400)	+

(Basic medium used was of the Wickerham's formula)

As indicated in the above table, *Wickerhamia fluorescens* requires biotin for the growth and other seven vitamins are not so essential. It seems that this has somewhat extensive adaptability on media. In the case of sporulation, the carrot agar (Soneda, 1955) was employed as pre-culture and malt extract agar, golodkova agar oat-meal agar and the other sporulation media were selected. As a result, oat-meal agar was most effective for the spore-formation. The abundant production of riboflavin was obtained on malt agar, carrot agar and rice-bran extract agar, but markedly reduced on synthetic medium as far as tested.

Optimum temperature for growth was 30°C. At 37°C on malt agar, it could not grow regularly.

Germination of ascospores can hardly be found in ordinary cases. But, the present author observed germination of them in very few cases. (Fig. 3)

The ascospores produced granulated, filamentous and thin-walled cells.

By Dr. Wickerham's personal communication, *Wickerhamia fluorescens* assimilated the followings: Glucose, galactose, sucrose, trehalose (very weak), raffinose, D-glucosamine, ethyl alcohol, glycerol, adonitol, mannitol, sorbitol, potassium gluconate, calcium 2-ketogluconate, pyruvate, lactate, succinate and citrate.

Nitrate was not assimilated. Vitamins were required. It grew well at 5°C, but did not grow at 37°C. Starch was not synthesized. Growth was almost nil in the medium

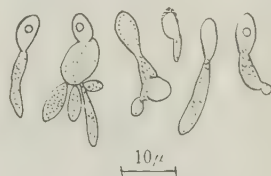


Fig. 3 Budding ascospores of *W. fluorescens*

of moderate osmotic pressure. Gelatin was not liquefied.

On detailed observations on the life-cycle of the present species are under investigation now and will be described in near future.

Summary

1) A new genus *Wickerhamia* was established as a member of Nadsoniae of Saccharomycetaceae, basing on *Kloeckera fluorescens*. On oat-meal agar developed abundant asci containing many ascospores of peculiar shape. Fermentation is positive and nitrate is not assimilated. It requires only biotin for growth.

2) This genus seems to be situated at the intermediate position between the members of Spermothoraceae and other members of Nadsoniae.

Acknowledgement

Grateful thanks are due to Dr. L.J. Wickerham, U.S. Department of Agriculture, for his valuable suggestion. Much of this work was also suggested by Dr. K. Kominami, Dr. Y. Kobayasi and Dr. K. Tubaki, to whom I owe thanks for their interests and helps.

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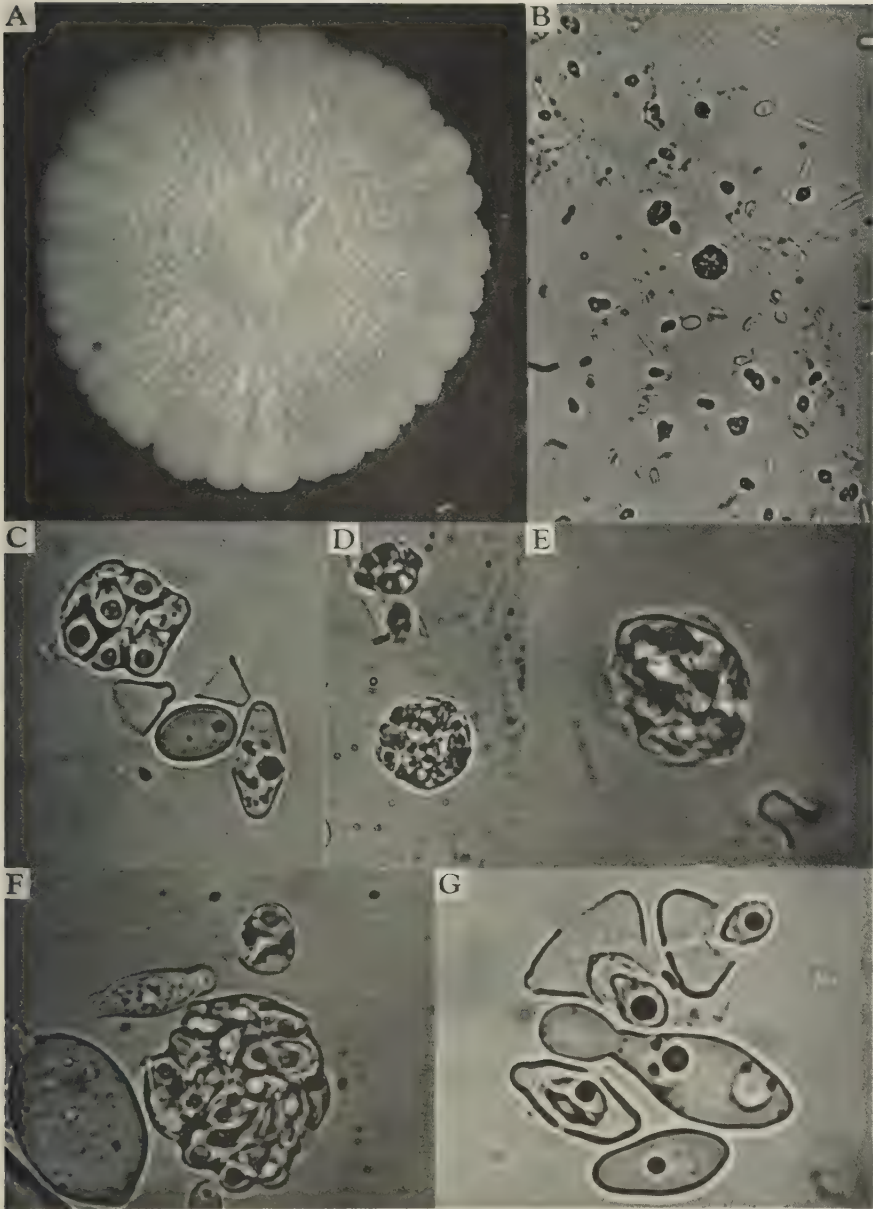


Plate 2 Photographs of *W. fluorescens*

A. Colony on malt agar plate 25. B. Various shapes of asci and ascospores. C. 8 ascospores in an ascus. D. Upper: 4 ascospores in an ascus. Lower: 16 ascospores in an ascus. E. 8 ascospores in an ascus and large cell. F. 16 ascospores in an ascus and large cell. G. Breaking down of three asci and budding vegetative cell.

On the Japanese Aquatic Hyphomycetes. Scum & Foam Group, referring to the preliminary survey of the Snow Group.*

By Keisuke TUBAKI

椿 啓介：日本産水生不完全菌類の研究．水泡生群及び雪表群（予報）

In an earlier paper (Tubaki, 1957), the present author described fourteen species of the aquatic Hyphomycetes from Japan and it was found that these members are quite common in Japan. The work recorded here is a continuation and amplification of the study of these fungi. After the excellent paper "Aquatic Hyphomycetes of decaying alder leaves" by Ingold (1942), many investigations on the aquatic Hyphomycetes have been carried out by Ingold in England, Ireland, Switzerland & Nigeria, by Ranzoni in California, by Nilsson in Sweden, by Suzuki and Nimura in Japan. In addition to the survey on aquatic members which develop on the submerged decaying leaves, the presence of spores of these fungi were reported in the foam and scum by Ingold (1956, 1959), Nilsson (1958) and Dixon (1959).

During 1958 and 1959, the present author examined many samples of foam & scum from nearly forty rivers and streams in Japan and found various aquatic spores in them (pl. 3-A, B). These spores in the rushing streams were collected directly by the plankton-net (pl. 3-C).

Further, as to know the ecological relation of these aquatic spores to the other non-aquatic condition, the precise examination of misty spray of water-fall and the remaining snow were also carried out (pl. 3). In collecting, samples of foam & scum were scooped up carefully into the sterilized stylol-tubes from the streams; the spray of water-fall was caught on the ordinal glass-slides on which 3% agar was coated; the planktonnet was prepared by 60 cm long and 30 cm in direct line, fixing the small stylol-tube to the base and drawn for 10 to 30 minutes in the rushing stream; the remaining snow was also taken into the stylol-tubes.

Foam & scum group : As pointed out by Ingold (1956, 1959), Nilsson (1958) and Nilsson by Dixon (1959), in foam and scum, conidia of aquatic Hyphomycetes were found in abundance. Nearly fifty samples of foam and scum were collected by the present author and examined directly under the microscope. The following fourteen

* This investigation was supported in part in a grant from the Committee of the Scientific Research of Yamagata Prefecture.

species were recognized in which four are newly found in Japan.

Alatospora acuminata Ingold (Fig. 1, A)

Trans. Brit. Mycol. Soc. 25 : 381(1942) ; Ranzoni, in Farlowia 4 : 377 (1953) ;
Tubaki, in Bull. Nat. Sci. Mus. 41 : 250(1957) ; Nilsson, in Sv. Bot. Tidsk. 52 :
307 (1958).

This species is also one of the very common scum-fungi and could be found in almost all samples.

Anguillospora crassa Ingold (Fig. 1, L)

Trans. Brit. Mycol. Soc. 41 : 367(1958).

This species was found four times in scum and foam in spring and summer. The conidia are characteristically S- or L-shaped and easily be recognized by these sinuous shape.

Conidia many septated, 160-200 μ long, 11-15 μ wide in the middle portion tapering to 5-8 μ at ends, hyaline. No conidiophore was found. The conidia of the present fungus differ slightly from that of Ingold in the narrower both ends and in the less-constriction at septa, but other features fit to the description.

Anguillospora longissima (Sacc. et Syd.) Ingold (Fig. 1, G)

Ingold, in Trans. Brit. Mycol. Soc. 25 : 402 (1942); Ranzoni, in Farlowia 4 : 361 (1953); Tubaki, in Bull. Nat. Sci. Mus. 41 : 252 (1957); Nilsson, in Sv. Bot. Tidsk. 52 : 294 (1958).

This species was already found in Japan and not so common in scum and foam.

Articulospora angulata Tubaki (Fig. 1, N)

Bull. Nat. Sci. Mus. 41 : 252 (1957).

This peculiar shaped spores were found three times in scum samples in spring. As far as I know, this fungus was not found in the other countries after I described from Japan, so this species seems to be restricted ecologically in Japan.

Articulospora inflata Ingold (Fig. 1, E)

Trans. Brit. Mycol. Soc. 27 : 35 (1944); Nilsson, in Sv. Bot. Tidsk. 52 : 296 (1958).

This species was found five times in scum and foam, and is new to Japan.

The conidia, the aleuriospore-type, consisting of main stalk and three divergent arms. Main stalk, 60 \times 3.5 μ , with slightly inflated uppermost part; one of the three arms which developed from the inflated portion of the main stalk not constricted at base, 115 \times 3 μ ; other two arms 100-130 \times 3 μ , constricted at each base.

Articulospora tetraccladia Ingold (Fig. 1, D)

Trans. Brit. Mycol. Soc. 25 : 376 (1942); Ranzoni, in Farlowia 4 : 364 (1953); Tubaki, in Bull. Nat. Sci. Mus. 41 : 253 (1957); Nilsson, in Sv. Bot. Tidsk. 52 : 294 (1958).

This species was already found in Japan and not so common in scum and foam.

Clavariopsis aquatica De Wild (Fig. 1, C)

Ann. Soc. Belge Microsc. 19 : 197 (1895); Ingold, in Trans. Brit. Mycol. Soc. 25 : 353 (1942); Ranzoni, in Farlowia 4 : 370(1953); Nilsson, in Sv. Bot. Tidsk. 52 : 300 (1958); Tubaki, in Bull. Nat. Sci. Mus. 41 : 255 (1957).

This species is one of the fairly common aquatic fungi in foam and scum in Japan.

Heliscus aquaticus Ingold (Fig. 1, J)

Trans. Brit. Mycol. Soc. 25 : 359 (1942); Ranzoni, in Farlowia 4 : 376 (1953); Nilsson, in Sv. Bot. Tidsk. 52 : 305 (1958).

This species seems fairly common one in scum and foam, and was newly found in Japan.

Conidia clavate, two-celled, often three-celled, with three short divergent arms, 28–37 μ long, hyaline.

Lemonniera aquatica De Wild (Fig. 1, B)

Ann. Soc. Belge Microsc. 18 : 143 (1894); Ingold, in Trans. Brit. Mycol. Soc. 25 : 343 (1942); Ranzoni, in Farlowia 4 : 378 (1953); Nilsson, in Sv. Bot. Tidsk. 52 : 307 (1958); Tubaki, in Bull. Nat. Sci. Mus. 41 : 260 (1957).

This species was already found in Japan and was found few times in scum and foam, but not so common.

Tetraccladium marchalianum De Wild (Fig. 1, H)

Ann. Soc. Belge Microsc. 27 : 34 (1883); Ingold, in Trans. Brit. Mycol. Soc. 25 : 369 (1942); Ranzoni, in Farlowia 4 : 366 (1953); Nilsson, in Sv. Bot. Tidsk. 52 : 296 (1958).

This species was found twice in scum and foam in spring though this is abundant in Britain (Ingold, 1959), California (Ranzoni, 1953) and Sweden (Nilsson, 1958).

Conidia, the aleuriospore-type, consisting of a main part, 25–30 \times 1–2 μ at base, widening to 3–5 μ at the upper part where a knob, 5–6 \times 6 μ , is formed. Below the knob, three arms develop. Two of them are 40–45 \times 2 μ , the third one is wider, 10–12 \times 2–3 μ and has a branch, 20–25 \times 2 μ .

This beautiful spore is clearly differs from that of *T. setigerum*, already reported, and from *T. maxilliformis* in having oval knob at the apex of main stalk.

Tetracadium setigerum (Grove) Ingold (Fig. 1, I)

Ingold, in Trans. Brit. Mycol. Soc. 25 : 369 (1942); Ranzoni, in Farlowia 4 : 366 (1953); Tubaki, in Bull. Nat. Sci. Mus. 41 : 261 (1957); Nilsson, in Sv. Bot. Tidsk. 52 : 298 (1958).

This species was already reported from Japan, and was found several times in this study in the samples of the plankton-net in northern parts of Japan.

Tetrachaetum elegans Ingold (Fig. 1, F)

Trans. Brit. Mycol. Soc. 25 : 337 (1942); Ranzoni, in Farlowia 4 : 371 (1953); Tubaki, in Bull. Nat. Sci. Mus. 41 : 261 (1957); Nilsson, in Sv. Bot. Tidsk. 52 : 300 (1958).

This species was already found in Japan and was found several times in the scum-samples in spring. This fungus seems to be one of the common species in scum and foam.

Tricladium splendens Ingold (Fig. 1, M)

Trans. Brit. Mycol. Soc. 25 : 385 (1942); Nilsson, in Sv. Bot. Tidsk. 52 : 299 (1958).

This fungus has been observed only once in scum in spring and newly found in Japan. By Nilsson (1958), this species seems to vary very much in size.

Conidia consisting of a slightly bent and septate main stalk, $45 \times 3 \mu$ at the middle part, from which two divergent and septate arms develop, $22-25 \mu$ long, 4μ at the widest parts, tapering to the apices, constricted at each base, hyaline. These two arms develop from the neighbouring cells.

Valicosporium elodae Kegel (Fig. 1, K)

Ber. d.d. Bot. Ges. 24 : 213 (1906); Ingold, in Trans. Brit. Mycol. Soc. 25 : 394 (1942); Tubaki, in Bull. Nat. Sci. Mus. 41 : 264 (1957); Nilsson, in Sv. Bot. Tidsk. 52 : 307 (1958).

Few conidia of this species were found in the scum in spring though not had been found in scum in Sweden (Nilsson, 1958); already reported from Japan.

In addition to the above members, there are a number of the aquatic Hyphomycetes belonging to unidentified species. However, as claimed by Ingold (1959), it is hopeful to refrain the erecting new taxa on the basis of only spores until the precise examina-

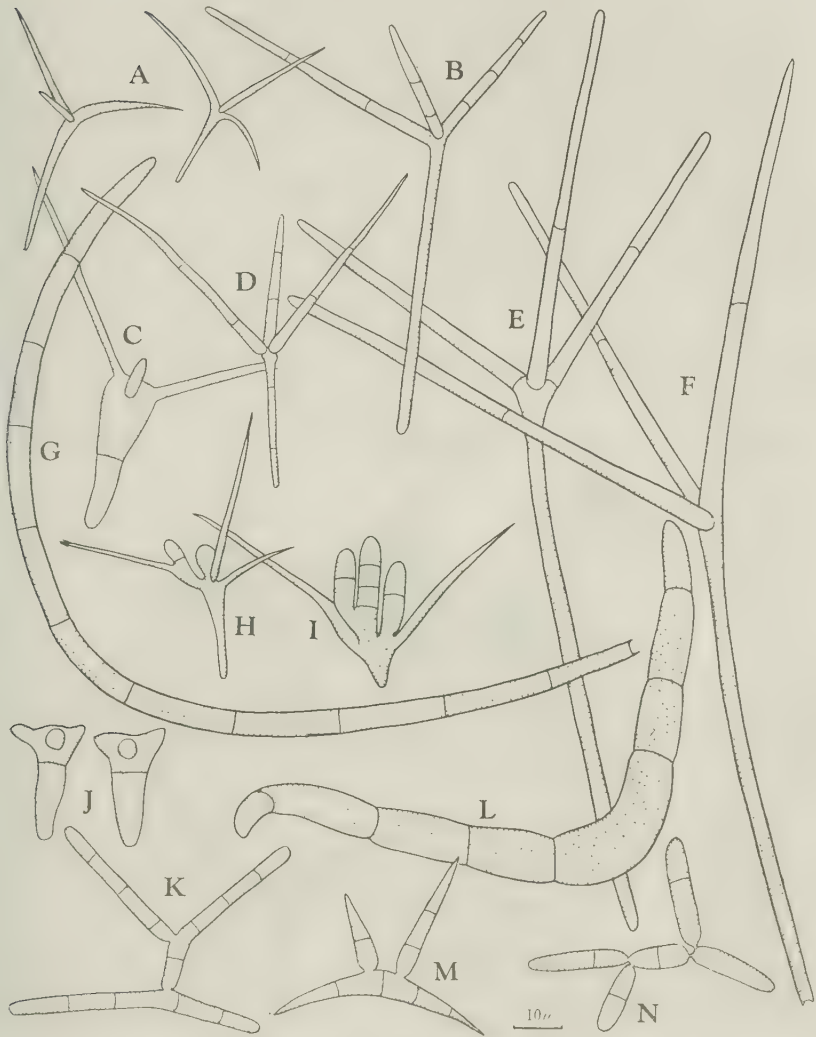


Fig. 1. Conidia found in foam & scum.

- A. *Alatospora acuminata* B. *Lenonnierea aquatica* C. *Clavariopsis aquatica*
D. *Articulospora tetracladia* E. *Articulospora inflata* F. *Tetrachactum*
elegans G. *Anquilospora longissima* H. *Tetracladium marchalianum* I.
Tetracladium setigerum J. *Heliscus aquaticus* K. *Valicosporium elodae* L.
Anquilospora crassa M. *Tricladium splendens* N. *Articulospora angulata*

tion is done on these sporulating mechanism under culture.

Conidia of unidentified species were shown in Fig. 2. Fig. 2 A could be seen in the scum of little stream (Nagano Pref. and foot of Mt. Iide, Yamagata Pref.) in abundance. Shape of the septate conidia approaches to the members of *Anguillospora*, but all conidia bent twice; foam an angle of nearly 180° at the first bending and nearly 90° at the second. It will be interesting to see the full stage of the conidial development. Fig. 2 B shows tetradiate spores which were found in several in the scum of small streams (Nagano Pref.) and resembles those of *Valicosporium elodae*, but differs from it in the acute points of all arms. Moreover, differs in the septation of arms of the latter species; those of the present fungus (4-) to (10-) pseudoseptated. The remarkable conidium shown in Fig. 2 C was obtained three times in the different streams and rivers of central and northern part of Japan in spring. These conidia near those of *Dendrospora erecta*, but clearly differ in slightly constricted septa and also in having knob-like cells at the upper part of main stalk. Many septated

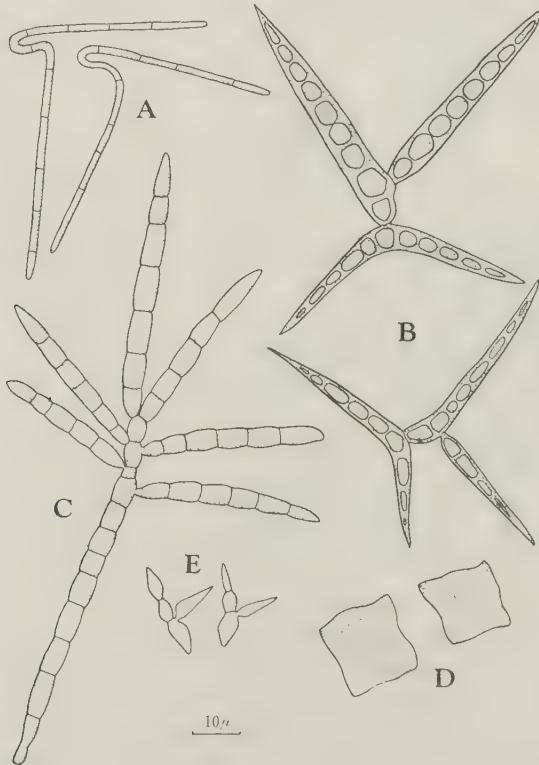


Fig. 2. Conidia of unknown species found in foam & scum.

arms, constricted at each septum, develop in several from the oval knobs or oval cells; each arm never branches more in contracting to those of *Dendrospora*. Few conidia, shown in Fig. 2D, were found in a foam-sample and near the tetrahedral conidia of *Margaritispora aquatica* in shape and also in containing the glycogen-vacuoles, but differ in larger size, $21-23 \times 20-22 \mu$. Conidia of Fig. 2E show those of unknown species.

In general, as indicated by Ingold (1959), scum and foam may act as a good spore-trap and contain many large number of the spores of aquatic Hyphomycetes together with those of terrestrial fungi. Plankton-net, previously described, is also useful for gathering of these spores, especially in the case of the rapid current. By this net, many spores can be collected (Pl. 3-C).

Detailed data on the habitat and the season of the collecting are summarized as follows.

species	Frequency	Habitat	Season
<i>Alatospora acuminata</i> Ingold	+++	scum & foam, by net	whole year
<i>Anguillospora crassa</i> Ingold*	++	scum & foam, by net	spring, summer
" <i>longissima</i> (S. et S.) Ingold	+	scum & foam	whole year
<i>Articulospora angulata</i> Tub.	++	scum & foam, by net	spring
" <i>inflata</i> Ingold*	+	scum	spring, summer
" <i>tetracladia</i> Ingold	+	scum & foam	spring to autumn
<i>Clavariopsis aquatica</i> De Wild	++	scum & foam	winter, spring
<i>Heliscus aquaticus</i> Ingold*	+++	scum & foam, by net	spring, summer
<i>Lemonniera aquatica</i> De Wild	++	scum & foam	spring
<i>Tetracladium marchalianum</i> De Wild	+	scum	spring
" <i>setigerum</i> (Grove) Ing.	++	foam, by net	spring to autumn
<i>Tetrachaetum elegans</i> Ingold	+++	scum & foam, by net	spring
<i>Tricladium splendens</i> Ingold*	++	scum	spring, summer
<i>Valicoporum elodae</i> Kegel	++	scum	spring

* newly found in Japan

Frequency.....+ : very few; ++ : fairly common; +++ : very common.

Spray of the water-fall

In July of 1959, the microflora of misty spray of the water-fall was examined by the above described method in Biwa Fall, Mt. Takao, near Tokyo. The formerly described 25 trapping slides were settled on the edges of the linn-rock, distant nearly 2 meters from the falling. The fall is rather of the small scale, filling the misty

spray inside of the linn, but no visible splash of water can get on the slides. The fall flows from the stream through a scarp covered with coppice, and water temperature is 9°C at the flowing. After the exposure of the slides for 30 minutes, they are removed and examined microscopically. Among them, on the six slides, spores of aquatic Hyphomycetes can be seen; these spores could be counted accurately and appeared, in number, between one and five in one slide.

Species examined are as follows :

- Anguillospora longissima* Ingold
- Articulospora tetracladia* Ingold
- Alatospora acuminata* Ingold
- Tetrachaetum elegans* Ingold

Above observation clearly demonstrates that these spores may detach from the surface of water and may fly into the air through the air-current, though this ecological course might be expected already by the other investigators. As regards the dispersal of these fungi, we must pay ecological attention to the another point in addition to the dispersal by water itself. These aquatic spores may flow and remove easily even by the minute movement, so it can be speculated that these spores may fly by even the minute air-current through the misty-spray of the fall. The characteristic spore-shape such as tetra- or filiform or sigmoid may serve not only for the dispersal in water but the flying up in the air. The spray of the water-fall and the dried substrate to which foam and scum attached may serve as the air-base for these fungi.

Remaining snow group

Snow as a habitat of microorganisms has only been studied mostly from the viewpoint of cryoxenous algae.

Few months ago, I had an opportunity of examining the remaining snow collected by Dr. H. Fukushima, Yokohama Municipal University, who is a phycologist and studying long times the members of the Cryophytes. It is of much interest to note that the remaining snow contained often fairly rich spora composed of the spores of terrestrial or aquatic Hyphomycetes. Of course, terrestrial spores in the remaining snow would be derived from air as already described by Kobayasi & Fukushima (1952), however, some spores of aquatic Hyphomycetes can also be seen by the precise examination of the melted snow. Then, during this spring and early summer, nearly sixty samples of the snow which were taken into the small tubes from the surface were gathered from foot or side of Mt. Asama, Mt. Tanigawa, Mt. Asahi, Mt. Iide and Ozegahara, all are situated at the central or northern part of Honshu, Japan.

In the tubes containing melted snow which are originally dirty white or almost

blackened, grayish or blackish precipitations appeared, and these precipitations were taken on the slides and examined carefully. Because each snow-sample is fairly restricted in proportion to the whole snow-surface and each contains only that of few centimeter-square, number of the aquatic spores in a sample is very few differing from that in the scum or foam which act as the trap of the spores. However, often the spores can be seen in abundance in a snow-sample, especially of that collected in the forest near the river. The number of spores seems for me not to be influenced by the kind of the tree of the forest and also not parallel with the turbidity of the melted snow. For an example, sample of clean snow contained few of these spores; on the contrary, one of the blackened snow contained only *Chionaster nivalis*, a typical cryoxenous alga. But, how do these aquatic spores can get on the snow? The presence of these aquatic spores on the surface of the remaining snow will indicate that these spores would be arrived to the snow by flying down like the common air-borne fungi from the stream or lakes as previously concerned. The presence of many spores on the snow of the forest will be resulted from the trapping of these spores by twigs or leaves. On the contrary, in samples of the snowy valley, rocky environment of Mt. Tanigawa, these spores were very few or almost lacking (Pl. 3-D). Those fungi found in the remaining snow were shown in Fig. 3.

In Fig. 3 A, an unidentified species was found on the snow in Ozegahara (June, 1959) which resembles those of *Ceratosporium*, but seems to be aquatic spores. *Articulospora tetracladia* Ing. (Fig. 3 B) and unidentified species (Fig. 3 D) were found on the snow in Mt. Hodaka, nearly 1600 meters over the sea-level (May, 1959). A fungus, shown in Fig. 3 D, was collected in the Mt. Hodaka's snow and seems to belong to *Tridentaria*, but is question whether aquatic or not. Pluriseptate conidia, shown in Fig. 3 E, were collected in Ozegahara and will be those of *Camposporium*. Like the above, conidia of Fig. 3 O, found in the snow in Mt. Asahi, is clearly a member of *Camposporium*, probably be *Camposporium antennatum* Harkness; this will be same with those in Nigeria (Ingold, 1959, fig. 2 e) and in Ghana (Dixon, 1958, Fig. 1 c). The curious conidium in the snow of Ozegahara, shown in Fig. 3 F, is probably be same with that in England (Ingold & Ellis, 1952, Fig. 1 h) and in Sweden (Nilsson, 1958, Fig. 12 c). Nilsson thought that the spore is to be a complex of conidia of *Trichocladium splendens*, however, his expectation seems not so reasonable for me; the findings of same spores in the three far distant countries, England, Sweden and Japan, will indicate to some extent that this spore will be an unknown aquatic conidium. A tetradial spore, shown in Fig. 3 G, were found in two snow samples of Mt. Tanigawa and Mt. Asama, and will be same with those in Ghana (Dixon, 1959, Fig. 1 a) in the stream flora. Both conidia of Ghana and Japan may be an aquatic spores of an unknown species. A remarkable spores, shown in Fig. 3 H, will be that

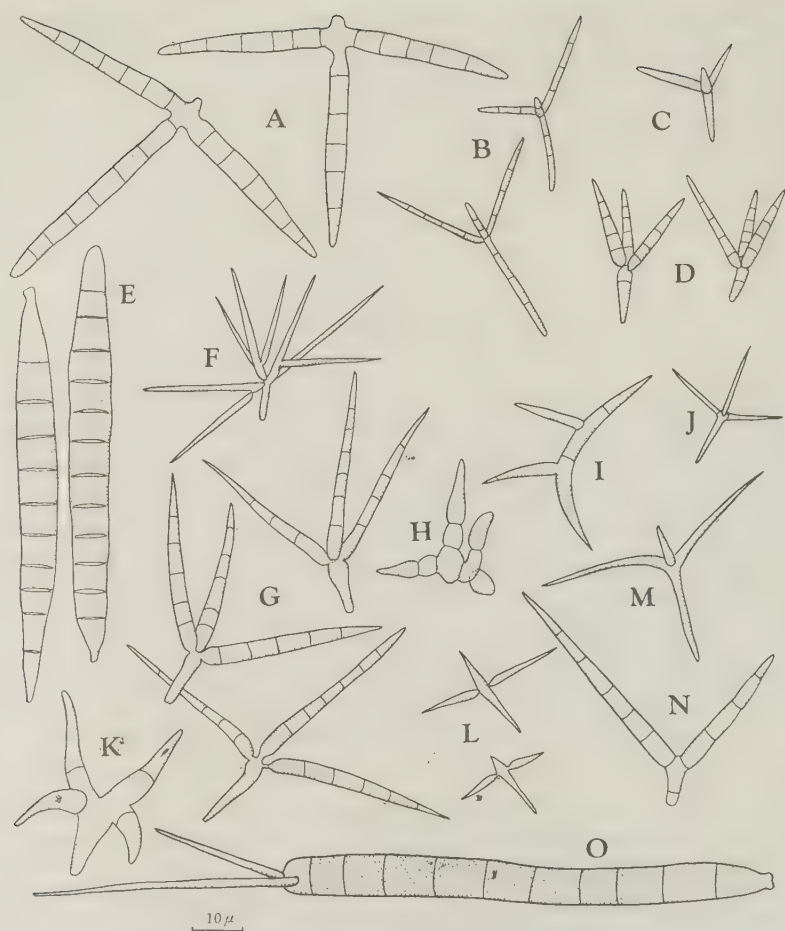


Fig. 3. Conidia found on the remaining snow.

- A.C.F.G.L. conidia of unidentified species. B. *Articulospora tetracladia*
 D. *Tridentaria* sp. E. *Camposporium* sp. H. *Tripospermum* sp.
 I. *Tricladium angulatum* J. *Heliscus* sp. K. *Campylospora chaetocladia*
 M. *Alatospora acuminata* N. *Ceratosporella* sp. O. *Camposporium antennatum*

of young stage of *Tripospermum*; a conidium figured by Ingold & Ellis (1952, Fig. 1 g) also seems to be a young stage of same fungus. Conidia in Fig. 3 I resembles that of *Tricladium angulatum* Ingold and was found once in the snow-sample, Asahi Nat. Park. An unknown species of *Heliscus* was found in the snow-sample in Asahi Nat. Park (Fig. 3 J) in fairly abundance. Though none of *Campylospora*-species has ever been found in Japan, a campylosporous spore-primordium of *C. chaetocladia* Ranzoni was found in a snow-sample in Mt. Asama (Fig. 3 K). Conidia in Fig. 3 L are those

of unknown fungus and could be found fairly in abundance in the Ozega-hara's snow-samples. Several conidia of *Alatospora acuminata* were found in the snow-samples in Asahi Nat. Park (Fig. 3 M). Few two-armed conidia (Fig. 3 N) were found in the snow-samples in Mt. Iide; this seems to be that of *Ceratosporella*-species, though it is questionable whether aquatic or not.

Furthermore, few samples of dirty scum were collected from the small pool into which the snow melted and flowed, where is originally dry and the pool disappears after the snow melts completely (Pl. 3-E, F). In these samples, the liberated spores of *Alatospora acuminata* Ing., *Articulospora tetracladia* Ing., *Anguillospora crassa* Ing. and those of figured in Fig. 3 G. were examined. The wet decaying leaves which were collected under these melting snow (Pl. 3-E) were put in the shallow dishes with clean water, then the following three species were found growing out from the leaves. This suggests that these leaves seem to be infected by the aquatic spores which fall down directly from the air or from the snow as it melts.

Tetrachaetum elegans Ingold

Lemonniera aquatica De Wild

Articulospora tetracladia Ingold

Of course, many terrestrial fungi can also be found on the snow (Fig. 3), but it is interesting that such helicosporous or phragmosporous fungi which prefer somewhat wet condition can be seen on the snow-surface.

As the above researches were held during only one snow-season, 1959-1960, further observations are necessary. This point is being investigated.

Discussion

As already pointed out by Ingold and Nilsson, the group of aquatic Hyphomycetes have a world-wide distribution and can be found during the whole year in nearly all waters. However, though there are certainly some still unknown species to be found in lake, pool, rapid or stagnant stream and scum & foam, most of those fungi found in microscopic-field are rather common and belong to the known species. This would strongly suggest that we are now in a position to make clear the following two problems besides of the examination of the flora of aquatic Hyphomycetes. The first problem is deduction of function of spores from such tetra-radiate form in water. This problem is being investigated by most ingenious method by Dr. J. Webster (Ingold, 1959), so this will come to be a conclusion in near future. The second problem is to trace the dispersal of the aquatic spores ecologically, especially to know the route of these spores going up to the higher area or to upper stream.

In the present paper, an account is given on collections of the aquatic spores from the foam & scum in water, and then a way of the dispersal of these fungi was found

by the precise examination of the misty spray of the water-fall. I also tried to make the preliminary report on the snow-group. The finding of the presence of these aquatic spores on the remaining snow also add the interesting information to the flora and the distribution of them in nature.

As already pointed out by Nilsson (1958), these aquatic conidia are very resistant and endure being dried and frozen. Consequently it might be speculated ecologically that these aquatic spores on the remaining snow would be distributed from the lake, river, stream, water-fall, foam or scum by the air-current, then they will disperse through the thawing stream to the lower water-habitat as the snow melts in spring or summer. When, in autumn or winter, these spores get on the decaying leaves on which it snows, the course of the distribution will be same with the above. However, the above hypotheses still have some questionable problems and it is interesting to catch the flying spores in mountainous area or on fresh snow though it is a very risky biological method.

Outside the fungi, the tetrastrate reproductive unit was discussed in the brown seaweed *Sphacelaria* in its function of dispersal (Ingold, 1959). In addition to the above alga, we must to pay the attention to the cryoxenous organisms, *Chionaster nivalis* (Bohl.) Wille (Fig. 4 A-1, 2) and *Selenotila nivalis* Lagerh. (Fig. 4 B); both were already described in Japan (Kabayasi & Fukushima, 1952 a). The former also can be seen in nearly all snow-samples in the present study and characteristically with three to four arms; that is not still decided whether it better to belongs to alga or not. This tetrastrate stage did not germinate even on malt agar as far as tested. Like this, the latter organism is question in its systematic position and thought to be one of the Hyphomycetes by these authors. This consists of a short arm and two or

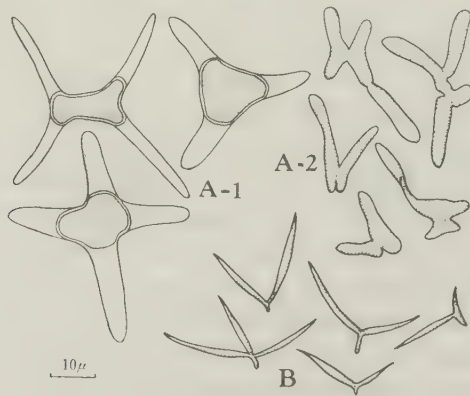


Fig. 4. Cryoxenous species

A-1, 2 *Chionaster nivalis*. (-1, typical; -2. immature stages) B. *Selenotila nivalis*

three slender, sickle-shaped, divergent arms (Fig. 4 B). The present author also found this reproductive unit in the snow and it seems for me to be a member of the aquatic Hyphomycetes.

In addition to them, *Raphidonema nivale* Lagerh. and *R. tatrae* (Kol) Vischer, both are called as cryoxenous algae and described in Japan by Kobayasi & Fukushima (1952 b), are very interesting for pointing out these shape of reproductive organs. The former resembles strikingly the conidia of *Anguillospora longissima* Ing. and those of the latter resembles *Flagellospora curvula* Ing. Both species seems for me as aquatic Hyphomycetes.

But it is better to refrain now from discussing the relation of the aquatic conidia to the cryoxenous algae on the bases of only shape of the reproductive unit until these organisms can examine under pure cultures. The discussion on the relationship awaits the precise examination in next spring.

In conclusion I would emphasize that the aquatic Hyphomycetes present not only in water or in foam & scum but in the misty spray of the water-fall and also on the surface of the remaining snow. It is fairly reasonable that there are certainly some striking aquatic conidia which can be known only from the snow.

Summary

An account is given of conidia of aquatic Hyphomycetes from foam & scum collected from central or northern part of Japan. Fourteen species are recognized and five unknown fungi are noted. The mycological examination of misty spray of the water-fall using the agar-slides shows the presence of four species in it. This indicates the possibility of the flying up of these spores into the air. The preliminary observations of the remaining snow from the viewpoint of the ecological base of aquatic Hyphomycetes show the presence of these fungi on it like those of cryoxenous algae.

Acknowledgements

The writer is indebted to Dr. Y. Kobayasi, Nat. Sci. Mus. Tokyo, for his many guidances during the investigations, and to Dr. H. Fukushima, Yokohama Municipal Univ., for his many suggestions on cryoxenous algae. Thanks are also due to Mr. D. Shimizu, Yonezawa Municipal Mus., for his many aids in collecting the samples. Mr. A. Seino, Mr. Y. Nomura, Miss. K. Suzuki and Miss. E. Ishikawa kindly offered the snow-samples to whom I owe many thanks.

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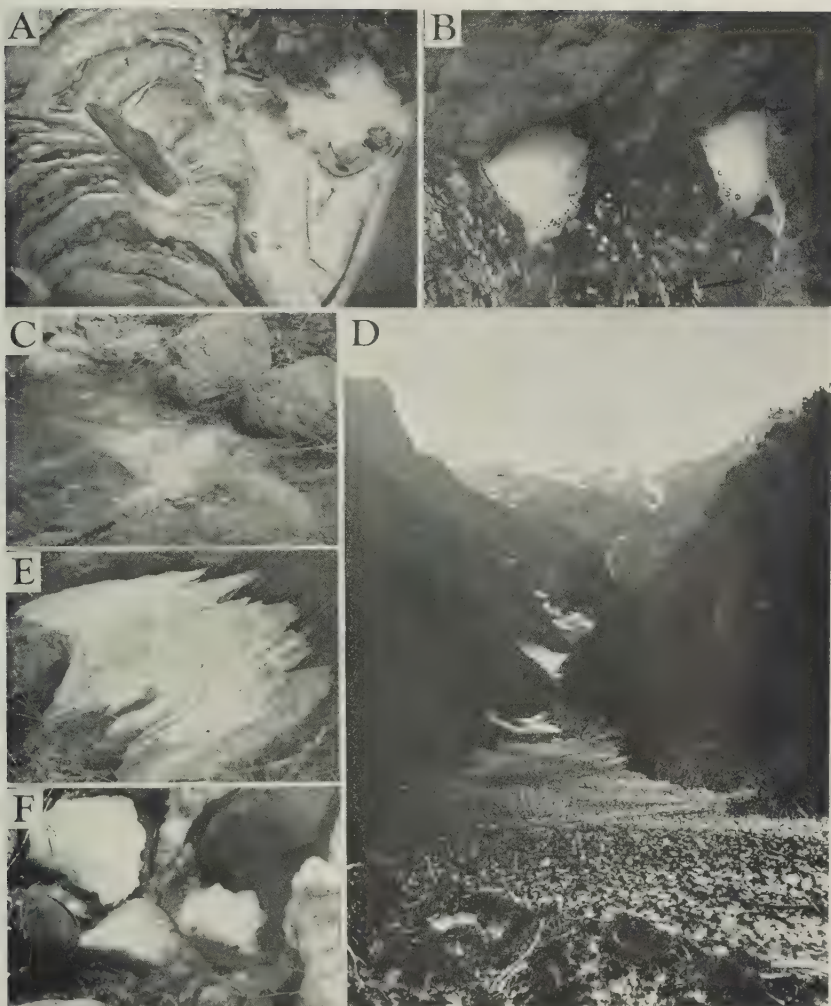


Plate 3. Several habitats of aquatic Hyphomycetes

A. Typical scum in small stream. B. Typical foam, growing at the side of the rock. C. Plankton-net, catching the fungi in the rushing stream. D. Snow valley in Mt. Tanigawa; rich aquatic fungi in the standing side and scant in the upper part. E. Remaining snow in Mt. Asahi, where it is originally dry. F. Dirty scum in the melting snow.

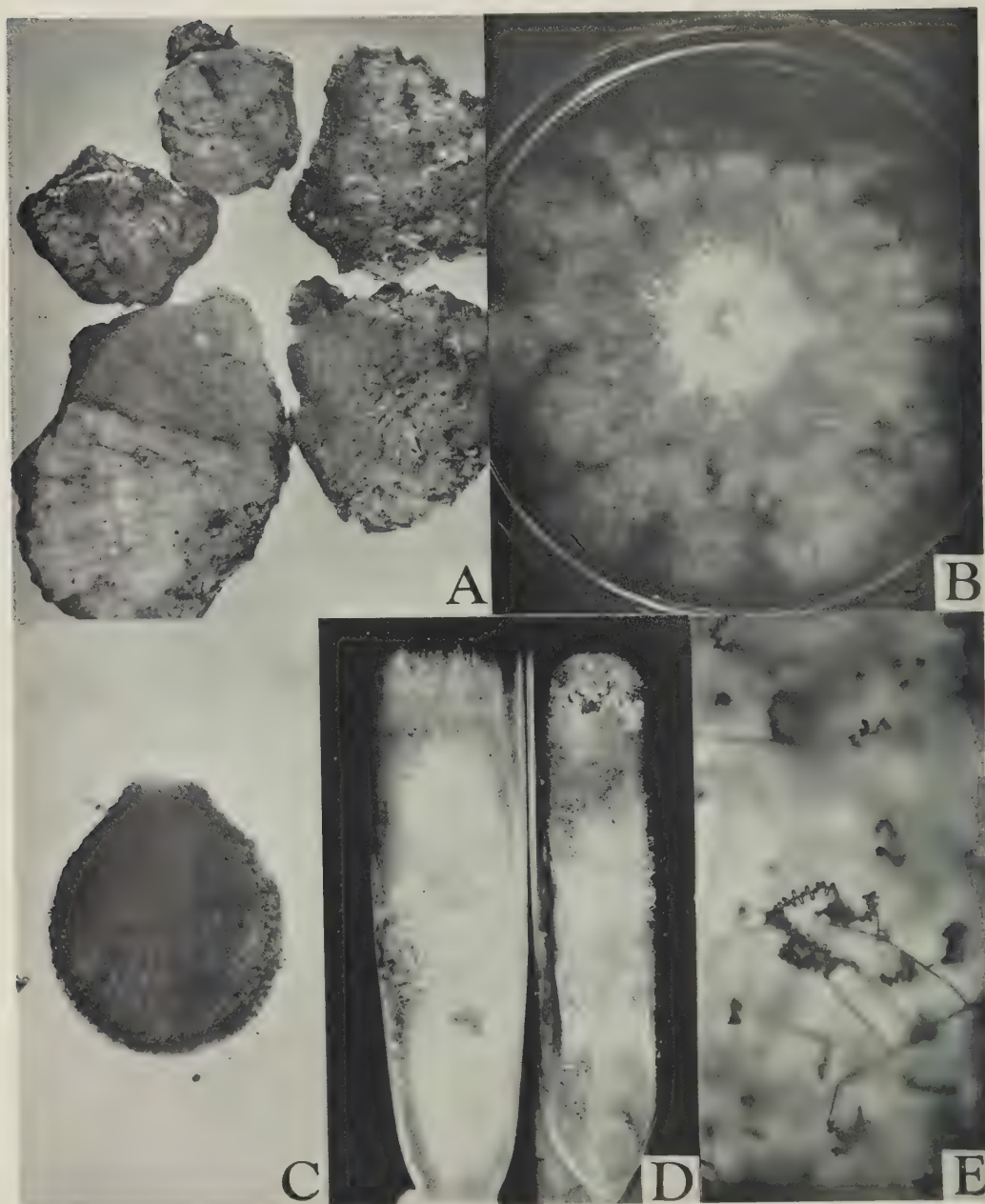


Plate 4. *Hypomyces trichothecoides*

A. Perithecia on the infected fruit-bodies of *Coriolus hirsutus* ($\times 1.3$). B. Colony on a malt-agar plate ($\times 1$). C. Perithecium ($\times 150$). D. Cultures on agar-slants showing the perithecial formation on upper surface (right, oat meal agar; left, malt agar). E. Conidial stage on malt agar ($\times 99$).

An undescribed species of *Hypomyces* and its conidial stage.

By Keisuke TUBAKI

椿 啓介：ヒポマイセスの一新種とその分生子世代について

In nature, the elucidation of natural relationships among the perfect and imperfect fungi awaits the discovery and study in pure culture. Thus, the present author has investigated the cultural stages of perfect fungi and discussed the relationships as already reported (1958), but no actual finding was yet obtained in the field in the members of *Hypomyces*.

This paper reports the finding of a new fungus with perfect character identical to the members of *Hypomyces* of which conidial stage is assignable to the form genus *Trichothecium*.

Hypomyces are characterized as parasitic or saprophytic upon higher fungi, especially upon Polyporaceae, and their conidial stages belong to the form-genera *Acrostalagmus*, *Dactylaria*, *Dactylium*, *Diplocladium*, *Didymocladium*, *Monosporium* and *Verticillium* as far as hitherto described by Seaver (1910), Petch (1938), Dingley (1950), Rogerson (1950), Munk (1957) and others. Chlamydospore-stage belongs, if present, to the form-genera *Sepedonium* or *Blastotrichum*. Most of them were already reported by the present author (1955).

The present fungus was originally found on the surface of fruit-bodies of *Coriolus hirsutus** in Yamagata Pref. in 1958. When collected, the surface of the host-fungus was covered entirely with the orange-red effuse subiculum with abundant perithecia

of *Hypomyces*. Pure culture of this fungus was obtained on malt agar after the germination of ascospores from the original perithecia. Then a curious shaped conidial fungus developed unexpectedly which does not belong to the any of known imperfect stages of *Hypomyces*.

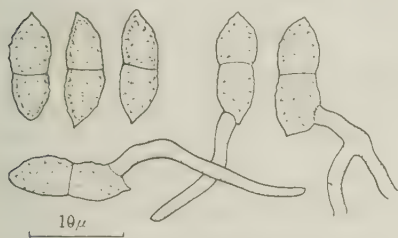


Fig. 1 Ascospores and their germinations

The growth of this fungus on malt agar is rather rapid, pure white at first and basipetal chains of conidia develop in a following peculiar manner. The primary conidium is produced obliquely from the apex of

* This host-fungus was kindly identified by Dr. K. Aoshima, Forest Experimental Station, Tokyo, to whom I owe many thanks.

end branch of conidiophore which branched verticillately, alternately or oppositely and delimited by a septum. Immediately below this septum, the branch swells to one side, forming a secondary conidium-primordium, which elongates, a septum then forms cutting off the secondary one which even has the primary conidium attached to it.

A swelling toward one side below the secondary conidium septum then forms the tertiary conidium-primordium, and development continues in this manner with the formation of successive conidia, each surmounted by the chain of younger conidia.

The conidia are continuous at first and then become two-celled, often three-celled.

As far as I know, fungus which forms didymous conidia in a chain by such a way is restricted only to *Trichothecium*-species; *Monosporium agaricinum* Bonord., *Cylindrophora apiculata* Tub. and *Mycogone rosea* Link also form the conidia by the same way, but the conidia of them are uni-celled and not in a chain (Tubaki, 1955).

In an earlier report of the present author (1958), I have arranged the conidia forming method of Hyphomycetes into nine sections which belong partly to Hughes' scheme (1953) and Section IX was newly added including *Trichothecium roseum* as typical fungus. I have also discussed the natural relationships between perfect and imperfect fungi from the viewpoint of the spore-forming mechanism in the same report and speculated that the members of Hyphomycetes which form conidia in such a manner of Sect. IX may connect with Hypocreales. The present report is a first one to account the group of Sect. IX and *Hypomyces*. Although the description of an imperfect stage belonging to *Trichothecium candidum* was published only once for *Hypomyces rosellus* by Tulasne (1865), it seems not be a true *Trichothecium*, but a *Dactylium*, perhaps *D. dendroides*, judging by the original figure as already discussed (1958).

Morphology

On malt agar slant at 25°, the fungus grows well, velvety or wooly, spreading with an irregular margin, pure white at first, then becoming yellowish orange. Conidial stage is of the *Trichothecium*-type, commonly developed in loosely tufts, pure white. Conidia catenulate in several to over 20 in a straight or curved chain, hyaline. The first formed conidia are long pyriform or clavate, acute at bases, two or three-celled; the subsequent conidia are ellipsoid, cylindrical or reniform, two-celled, often with the nipple-like projections at the points of the attachment. The perithecia are usually formed after two months on malt agar or oat-meal agar slant, gregariously at the upper parts of the slant, superficial or semi-immersed in the subiculum, globose or oval, yellowish orange to dark orange red. The asci are cylindrical, containing eight ascospores which arranged uniseriately. The ascospores are of that of typical *Hypomyces*, didymous, verrucose, same with that in the field; producing germ-tubes at one or both ends at 25° after 24 hours. In the characteristic orange-red color of the peri-

thecia of this fungus, it is of typical *Hypomyces aurantius*, but the size of ascospore is smaller. This fungus also differs from the nearby species, *H. fulgens* and *H. javanicus*, in many points after the treatments of the specimens (State Museum of Natural History, Stockholm)*. According to Dr. Rogerson's personal communication, my specimen also does not match with the species such as *H. polysporus*, *H. australiensis*, *H. cesatii*, *H. albidus*, *H. sulphureus* and *H. chromaticus*. Therefore, even in regard to the ascospore-size, the present fungus does not fit in any of the known species. Moreover, the conidial stage, a *Trichothecium*-type, is clearly different from those of hitherto described species. As already mentioned (1958), conidial stages of ascomycetous fungi, including the type of conidiophore and conidial development, must be considered just as important in indicating the natural relationships among them as those of the perithecia and their contents. In the case of the present fungus, the conidial stage is to be stressed in the taxonomy like the characters of sexual organs and the coloration.

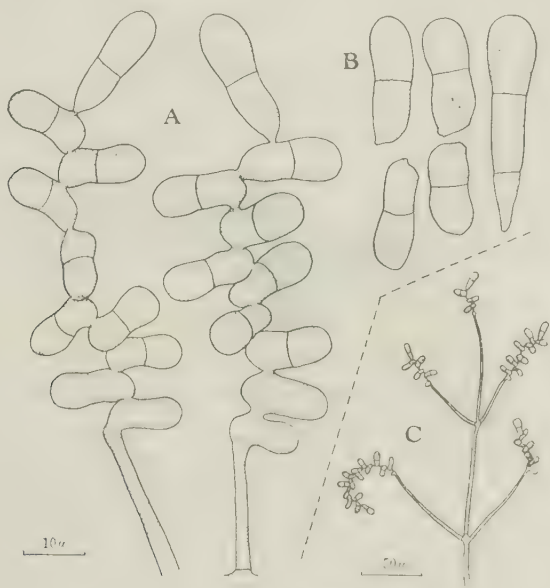


Fig. 2 *Hypomyces trichothecoides*

A. Successive formation of conidia B. Conidia C. Habit

On these bases a new species of *Hypomyces* is established.

***Hypomyces trichothecoides* Tubaki sp. nov.**

Subiculum effusae, rapide incrementes, aureae dein ochraceae, marginae

* These specimens were kindly offered by Dr. T. E. Hasselrot, the Curator of the Museum, to whom I wish to express many thanks.

floccosum. *Perithecia gregariae, superficialiae, globosae vel ovoideae, ostiolum papillatum*, $230-300 \times 250-370 \mu$, ochraceae dein sanguineae. Asci cylindricae, truncatae, $80-110 \times 4-5 \mu$. Ascospora uni-septatae, fusiformae, verrucosae, $14.5-16.5 \times 3.5-4.0 \mu$, hyalinae. Status conidiis: *Trichothecium*-forma. Conidiophora sympodialiter vel monopodialiter ramosa, $3.5-5 \mu$ crassa. Conidia primariae elongate pyriformia vel clavata, 1- vel 2-septata, basi acuta, $20-35 \times 5.5-7.0 \mu$; conidia continuatae, ellipsoidea vel reniforma, 1-septata, $14-25(30) \times 3.5-7.0 \mu$, hyalina.

Subiculum effuse, spreading over the entire surface of the host, light yellow and orange-red, often with a white margin; perithecia gregarious, crowded, superficial or semi-immersed in subiculum, globose or oval with slightly papillated ostiole, $230-300 \times 250-370 \mu$, golden yellow or orange yellow; asci cylindrical, with truncated ends, thin-walled, $80-110 \times 4-5 \mu$, eight obliquely uniseriate spored; ascospores fusiform, straight or slightly curved, didymous, slightly constricted at septa, verrucose, with highly granular contents, $14.5-16.5 \times 3.5-4.0 \mu$, hyaline. Conidial stage, the *Trichothecium*-type; conidiophores $3.5-5 \mu$ width, branched alternately or oppositely, bearing conidia from the apices obliquely or horizontally in a chain; terminal conidia two- or three-celled, long pyriform or clavate, acute at bases, $20-35 \times 5.5-7.0 \mu$, subsequent conidia ellipsoid, cylindrical or reniform, two-celled, $14-25(30) \times 3.5-7.0 \mu$, hyaline.

Hab. On fruit of *Coriolus hirsutus*, Neko Village, Asahi National Park, Yamagata Pref. (Sept., 1958). Type specimen was deposited in Nagao Institute.

Physiology

I. Carbon- and nitrogen-assimilation

As to know the favourable condition for the growth, the following assimilation-tests were carried out. The basal medium* used was adequate to support fair to good growth, provided that the sugar or nitrogen supplied was utilized. To this basal medium, the sugars were used as a rate that supplied 1% of carbon per liter and $(\text{NH}_4)_2\text{SO}_4$ (0.1%) was used as a sole source of nitrogen for the carbon-assimilation test. For the nitrogen-test, the nitrogen compounds were supplied 0.1% and glucose was used as a sole source of carbon. Cultures were kept at 25° for fourteen days. As the result of carbon-test, D-glucose, D-fructose, D-mannose, maltose, sucrose, cellobiose, trehalose, raffinose and soluble starch were good sources for the growth; D-galactose and D-xylose were utilized slightly; L-arabinose, lactose and sodium

* KH_2PO_4 1 g; MgSO_4 0.5 g; NaCl 0.1 g; L-histidine monohydrochloride 10 mg; DL-Methionine 20 mg; DL-tryptophane 20 mg; Biotin 2 μ g; Calcium pantothenate 400 μ g; Inositol 2000 μ g; Niacin 400 μ g; Para-aminobenzoic acid 200 μ g; Pyridoxine hydrochloride 400 μ g; Riboflavin 200 μ g; Thiamine hydrochloride 400 μ g; Distilled water 1000 ml; agar 20 g; pH was adjusted to 6.5

acetate were refused. As nitrogen, peptone and ammonium sulfate were good sources for the growth.

II. Carotenoid pigment

This fungus was inoculated on the sterilized fruit-bodies of *Coriolus hirsutus* and kept at 25° for nearly one month, then orange-red mycelium developed on all surface of the host. These colored mycelia were picked up carefully, then dried and powdered. Pigment was extracted by the method of Ellinghausen & Pelczar (1954), and methanol- and chloroform-extract were obtained. The materials thus extracted were examined spectrophotometrically in a Beckman DU Spectrophotometer as a spectrum band width of 5 m μ . The wavelength ranged from 400 to 700 m μ . Absorption peak is 475 m μ in methanol. This indicates that the orange-red pigment of the present fungus is consist mainly of carotenoid pigment.

III. Vitamin requirement

Because the growth of this fungus on such synthetic media as Czapek agar, is very markedly reduced as compared with that on malt agar, the effect of eight vitamins to the growth were examined. To the same prior basal medium, except for the vitamins, glucose (1%) and ammonium sulfate (0.1%) were added. The eight vitamin-compounds supplied are same with that of these in the prior basal medium. Burkholder's method was introduced which detects a deficiency by inability to grow in a medium of complete set of vitamins except for one. Failure to grow in a medium indicates a deficiency for the vitamin omitted. As a result, it appears that the present fungus requires only a thiamine for the growth; this is a thiamine-deficient fungus.

Summary

An undescribed orange-red species of *Hypomyces* was collected and examined under culture. The dried specimen of it approaches *Hypomyces aurantius* in all respects except for the smaller ascospores, but clearly differs from the latter species in a conidial stage, *Trichothecium*-type which is a first record as a conidial stage of *Hypomyces*. Therefore, a new taxon, *Hypomyces trichothecoides* is proposed. Perithecia develop on the semi-natural media, such as malt agar and oat-meal agar, in fairly abundance, and it is proved spectrophotometrically that the orange-red coloration of them is due to the carotenoid pigment. Tests for carbon- and nitrogen-assimilation are carried out. This fungus is thiamine-deficient.

Acknowledgment

Many thanks are due to Dr. Y. Kobayasi, National Science Museum, Tokyo, for his constant guidances and also to Mr. D. Shimizu, Yonezawa Municipal Museum, for many courtesies during the collecting of the fungus in the field. The writer is

also indebted to Dr. C.T. Rogerson, New York Botanical Garden, and to Dr. J.M. Dingley, Dept. Scientific & Indust. Res., Auckland, for their valuable suggestions.

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On the *Elaphomyces* found in Japan

By Yosio KOBAYASI

小林義雄：日本産エラフオミセス菌について

Ten years or more have elapsed since the writer began the collection of the members of *Elaphomyces* in Japan as the byproduct during the study of fungicolous *Cordyceps*.

The Elaphomycetous underground fungi were already described in 1742 with the generic name *Lycoperdastrum* by Haller, although the genus *Elaphomyces* was established far later in 1820 by Nees von Esenbeck. Since then, the monographic studies including the European species had been pursued by several mycologists such as Vittadini (1831), Tulasne (1851), Hesse (1894) and Hollós (1911). In 1891, Fischer enumerated 21 species of this genus and systematized them in Rabenhorst's Kryptogamen-Flora. In practical use, there are, however, some unreasonable treatments in it. Accordingly the monographic paper by C.W. Dodge (1929) should be taken as the most reliable and up-to-date literature, although *El. japonicus* Lloyd (1916) is omitted in it. The writer followed him in this study. It is very difficult and useless to discuss the specific definitions basing only on the above mentioned old literatures without reexamining their authentic materials. Once the writer tried in vain to culture purely *El. granulatus* by cutting the fructification and transferring a piece of peridium on malt agar plates. It seems to be difficult to culture such a mycorrhizal fungus.

In Japan, the members of *Elaphomyces* were found among coniferous forests (*Pinus*, *Tsuga*, *Thujopsis*, *Picea*, etc) as well as in deciduous forests (*Quercus*, *Betula*, *Fagus*, etc).

Imai enumerated eight Japanese species in his last paper (1943). Among them, *El. subvariegatus* is considered to be the same with *El. variegatus*. The writer enumerates, here, the following fourteen species, among which *El. anthracinus*, *El. reticulatus* and *El. muricatus* are new additions to the Japanese flora, and *El. asahimontanus*, *El. titibuensis*, *El. shinizuensis* and *El. neoasperulus* are new to science.

Subgenus *Scleroderma* Vitt.

Sec. *Ceratogaster* (Corda) Fr.

- 1) *Elaphomyces asahimontanus* Y. Kobayasi
- 2) *El. nopporensis* Imai
- 3) *El. anthracinus* Vitt.

Sec. *Ceraunion* (Wallr.) Fr.

Subsec. *Atrocorticis*

- 4) *El. miyabeanus* Imai

- 5) *El. nikkoensis* Imai
- 6) *El. titibuensis* Y. Kobayasi

Subsec. *Cerinocorticis*

Ser. *Variegatae*

- 7) *El. reticulatus* Vitt.
- 8) *El. variegatus* Vitt.
- 9) *El. muricatus* Fr.

Ser. *Homogeneae*

- 10) *El. fragilisporus* Imai
- 11) *El. japonicus* Lloyd
- 12) *El. granulatus* Fr.
- 13) *El. shimizuensis* Y. Kobayasi
- 14) *El. neosperulus* Y. Kobayasi

Since the publication of Dodge's paper, relatively few species have been added. Except for the four new species by Imai, those additional species are enumerated as follows.

- Elaphomyces appalachiensis* Linder from U.S.A. (1939).
- El. sulphureo-pallidus* Vacek from Czechoslovakia (1949).
- El. carbonaceus* Corner et Hawker from Singapore (1953).
- El. singaporensis* Corner et Hawker from Singapore (1953).

Thirty-seven total species including the writer's four new species, have the following distribution type.

Proper in Europe	15	Proper in N. America	2
Proper in Japan	9	Proper in Singapore	2
Common to Europe and N. America	3		
Common to Europe and Australia	1		

Common to Europe, N. America and Japan (almost cosmopolite in the Northern Hemisphere) 5

Judging from the distribution of *Cordyceps capitata* and *C. ophioglossoides*, there may be found some species of *Elaphomyces* as the host fungi of *Cordyceps* in Mexico, Eastern Siberia and China.

As the conclusion, this genus is considered to be confined to the temperate region of the Northern Hemisphere with some exception. On the contrary, the *Mesophellia* and its related primitive genus are distributed in the Southern Hemisphere, instead of the *Elaphomyces*.

In Japan, Several fungi were found in sterile condition in spite of the fully matured sizes. The gleba of these fungi was stuffed with elongate and brownish cell membrane, showing powdery appearance. Presuming from other characteristics than glebal

structures, they were identified as follows.

Elaphomyces granulatus as host of *Cordyceps* \times *jezoensisoides* and *C. valvatostipitata*

El. reticulatus as host of *C. ophioglossoides*

El. neoasperulus as host of *C. japonica* and *C. valvatostipitata*

It is interesting that all the fructifications of host of *C. valvatostipitata* were sterile.

Technical terms used in this paper are shown in Fig. 1.

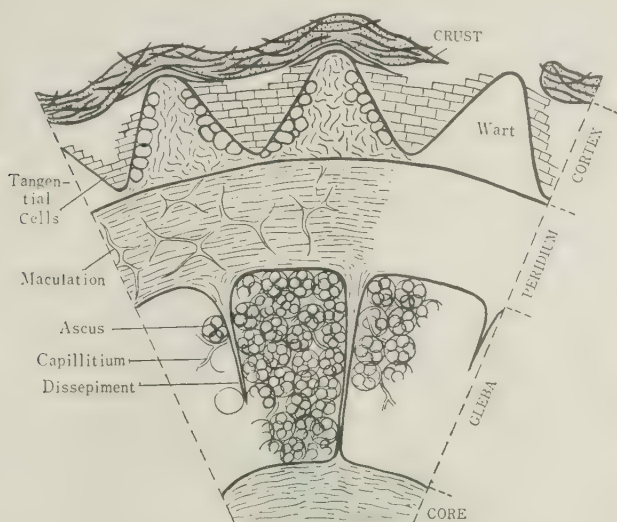


Fig. 1. Schematic figure of section of fructification (*Ceraunion*)

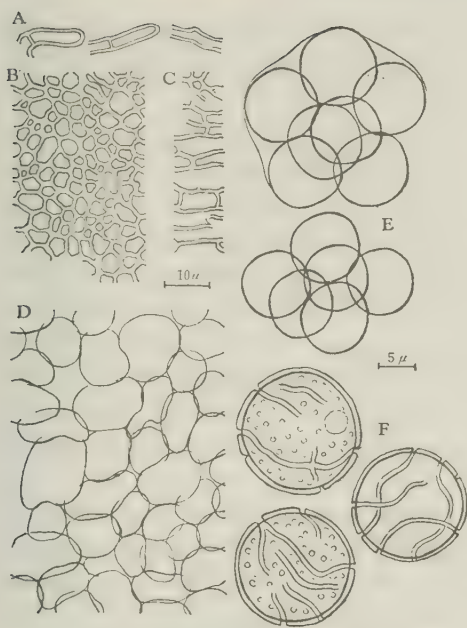
Key to the Japanese species

- 1 Cortex wrinkled, \pm fleshy, spores small, $>15\mu$Subg. *Malacoderma*
(not yet found in Japan)
Cortex smooth, verrucose, echinulate, spores large, $15-20\mu$ or more.....
.....Subg. *Scleroderma*.....2
- 2 Cortex smooth or nearly so, mostly carbonaceous... Sec. *Ceratogaster*.....3
Cortex verrucose to echinulate, \pm coriaceous.....Sec. *Ceraunion*.....6
- 3 Ascospores smooth, hyaline or light yellow.....[*El. leucosporus*]
Ascospores not smooth.....4
- 4 Ascospores sulcate, pale vinaceous.....*El. asahimontanus*
Ascospores echinulate.....5
- 5 Ascospores about 15μ , dark brown to black, peridium papery.....
.....*El. nopporensis*
Ascospores $17-20\mu$, cinereous brown, semitransparent. Peridium

- 1.5–2 mm, cottony-fleshy.....*El. anthracinus*
- 6 Cortex black or dark brown.....Subsec. *Atrocorticis*
Cortex yellowish, ochraceous, cinnamon or fuliginous.....
..... Subsec. *Cervinocorticis*
- 7 Warts large up to 1 mm high, irregularly distributed, largest
in the central part, spores 15–22 (commonly 20) μ , echinulate
..... *El. miyabeanus*
Warts small, 200–250 μ high, spores 25–30 μ8
- 8 Spines of spore finely fasciculate, seemingly verruculose.....*El. nikkoensis*
Spines Conspicuous.....*El. titibuensis*
- 9 Peridium marbled (maculated).....Ser. *Variegatae*
Peridium homogeneous.....Ser. *Homogeneae*
- 10 Warts flat, verrucose, small, 200–250 μ high.....*El. reticulatus*
Warts pointed or obtuse.....11
- 11 Warts small, 300 μ or more high.....*El. variegatus*
Warts large, prominent, 900–1,000 μ or more high.....*El. muricatus*
- 12 Warts low, not pointed, granulose, commonly 200–300 μ high.....13
Warts high, pointed, 400–750 μ16
- 13 Spores reticulate or verruculose.....*El. fragilisporus*
Spores echinulate.....14
- 14 Ascospores very densely beset with low spines (ca 1 mm),
coalescent in blocks. Peridium grayish red.....[*El. asperulus*]
Spines of spore frequently fasciculate, but never in large blocks.....15
- 15 Spines of spore short, up to 2 μ . Peridium vinaceous.....*El. japonicus*
Spines long, attaining 3–4 μ . Peridium white or grayish white
..... *El. granulatus*
- 16 Spores minutely papillate.....*El. shimizuensis*
Spores very densely set with low spines (1 mm), frequently
coalescent in blocks.....*El. neosperulus*

1. ***Elaphomyces asahimontanus*** Y. Kobayasi sp. nov.

Fructificationes globosae 5 mm in diam., superne partim encrustae cum particulis terrarum. Cortex cinereoso-niger vel ater, scaber irregulariter areolatus, sine verruca, sectione 125–155 μ crassus, nigritellus, carbonaceus, e cellulis elongatis crassiparietalibus (4–6 μ in diam.) dense contextus. Peridium 220–320 μ crassum, pallide fuscum, pseudoparenchymaticum. Textus inter peridium et glebam tenuissimus membranaceus pallide cinereoso ochraceus e hyphis 1–1.5 μ crassis tenuiparietalibus compositus. Gleba farinosa ochraceo-vinaceus, dissepimentis destitutis. Asci cum 6–8 sporis. Ascosporae

Fig. 2. *El. asahimontanus*

A. Superficial hyphae B. C. Tissue of cortex
D. Tissue of peridium E. Asci F. Ascospores

sphaericae $13.5\text{--}22\ \mu$ in diam., pallide vinaceae, glabrae vel obscuriter scabrae, irregulariter sulcatae episporis ca $1\text{--}1.5\ \mu$ crassis. Capillitia hyalina septata ramosa $1.5\ \mu$ crassa.

Fructifications globose 5 mm in diameter, partially encrusted with particles of earth, mycelial strang scanty. Cortex cinereous black, scabrous, irregularly areolate, without warts, in section $125\text{--}155\ \mu$ thick, almost black, carbonaceous, brittle, densely composed of thick-walled elongate cells ($4\text{--}6\ \mu$ in diameter). Peridium well differentiated from cortex, $220\text{--}320\ \mu$ thick, pale brown, pseudoparenchymatous. Hyphal tissue between peridium and gleba very thin, papery, pale cinereous-ochraceous (silvery white by naked eye) composed of $1\text{--}1.5\ \mu$ thick hyphae. Gleba powdery, ochraceous-vinaceous,

without dissepiments. Asci 6, 7, 8 spored. Ascospores sphaerical, $13.5\text{--}22$ (commonly 20) μ in diameter, pale vinaceous, smooth or obscurely scabrous, sulcate with irregularly running grooves, episporium ca $1\text{--}1.5\ \mu$ thick. Capillitia hyaline, $1.5\ \mu$ thick, septate and branched.

Hab. Underground of 3–4 cm depth, in mixed forests on the slope of small valley. Host of *Cordyceps delicatostipitata*. Yanabazawa of Owizawa, Yamagata Pref. (Sept. 13, 1958, Y. Kobayasi & D. Shimizu) Type (No. 52) in Herb. Nat. Sci. Mus.

This is near *Elaphomyces leucosporus*, differing, however, in spore sculpture. In peridial tissue mingled very slender hyphae which are hyaline and $1\text{--}1.5\ \mu$ thick. These may be the parasitic hyphae of *Cordyceps*.

2. *Elaphomyces nopporensis* Imai, in Trans. Sapporo Nat. Hist. Soc. 11: 36 (1929).

"Fructifications small globose or subglobose, 3.5–10 mm in diam. Crust forming loose mass around fructifications, mingled with blackish brown mycelia. Cortex dark or blackish brown, nearly smooth, but rough under the lens, very thin, 0.25 mm thick, rigid, not shrunken when dried. Peridium very thin, papery, clove-brown, easily separable from cortex when matured. Gleba blackish, powdery when matured, disse-

piments inconspicuous at maturity. Ascospores globose or subglobose, 15–17.5 (mostly 15) μ , thick walled, densely echinulate, dark brown to black. Capillitium yellowish, granulate”.

Nom. Jap. Kokuro-tutidango

Hab. Host of *Cordyceps jezoensis*. Nopporo, Hokkaido (by Imai). Known only from type locality. Type in Herb. Imai.

As shown from the above description by Imai, this is very near *El. anthracinus*. In future, this may be found to be conspecific with latter species. Now the writer considers this provisionally separable from *El. anthracinus* by the small fructifications and very thin cortex and peridium.

3. ***Elaphomyces anthracinus*** Vitt., Monogr. Tuberac. p 66 (1831); C.W. Dodge, in Ann. Myc. 27(3,4) : 165 (1929).

Syn. *Elaphomyces uliginosus* Hesse, Hypog. Deutschl. 2 : 67 (1894); Lange in Dansk Bot. Arkiv 16 (1) : 25 (1956).

Fructifications depressed globose, 18 \times 15 mm, without crust. Cortex black, somewhat glossy, without warts, finely scabrous by naked eye, finely and irregularly granulous under lens, 300–500 μ or more thick, homogeneous, carbonaceous, black, brittle fragile, easily separable from peridium, with irregularly denticulated surface, sclerenchymatous composed of brown-black (dark sepia), thick walled polygonal cells. Hyphae on surface of cortex irregularly and sparingly running, vinaceous-brown, somewhat thick walled, 3.5–4 μ thick, septate, sparingly branched. Peridium 1.5–2 mm thick, cottony carneous, pale vinaceous with 1 mm broad darker zone in the middle, composed of thin-walled, loosely woven, 1.5–2.5 μ thick, pale vinaceous hyphae. Gleba stuffed, brownish black, with pale-coloured hyphal zone in the middle; dissepiment slender, almost disappearing in adult stage. Ascospores sphaerical, 17–20 μ , cinereous brown, with low spines, semitransparent. Capillitia thin-walled, hyaline, 2.5–3 μ thick, branched. Odour unknown.

Hab. Host of *Cordyceps jezoensis*. Inego, Katta-gun, Miyagi pref. (Sept. 8, 1957, D. Shimizu).

Distr. Europe, North America (N. Carolina, Tennessee)

Except for ascospores, Japanese fungus was found surely identical in all details with the description by C.W. Dodge. According to his description and notes, and that of *El. uliginosus* by M. lange, the ascospores vary from almost smooth to slightly aspe-

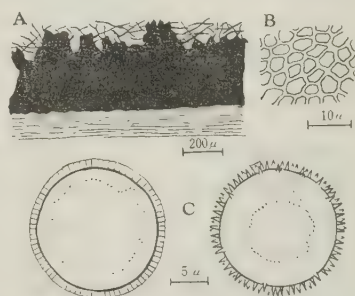


Fig. 3. *El. anthracinus*

A, B. Section of cortex
C. Ascospores

rate or more asperate.

4. *Elaphomyces miyabeanus* Imai, in Trans. Sapporo Nat. Hist. Soc. **11** : 35 (1929).

"Crust forming the loose or rigid mass around the fructifications with blackish brown mycelia. Fructifications subglobose, depressed or subovate, $15-30 \times 15-25 \times 15-20$ mm, surface more or less rough and provided on the apical or rarely on the basal part with warts. Cortex blackish brown or black, rigid, hard, 0.5 mm thick, not shrunk in drying, easily separable from the peridium when matured. Warts pyramidal with blunt apex, largest ones in the center, the size diminishing centrifugally, up to 1 mm high. Peridium grayish white with fine pinkish lines, more or less firm, 1-2 mm thick, becoming thinner and dull pinkish in colour when matured or dried. Gleba bluish gray at first, becoming blackish-brown. Core forming white cottony mass of hyaline hyphae in the center of fructification, from which dissepiments radiate as white sterile veins and their side branches forming many compartments, finally dissepiments and core disappear. Asci 8-spored. Ascospores globose or subglobose, with a central gutta, $15-22$ (mostly 20) μ in diameter, wall thick, yellowish at first and with radial fine striation, finally becoming blackish brown or black colour and densely echinulate. Capillitia white unevenwide, with many small guttae and provided with small hyaline crystals on the wall, at length becoming slender and hyaline."

Nom. Jap. Kuro-tutidango

Hab. Host of *Cordyceps jezoensis*. Known only from type locality. Nopporo, Prov. Isikari, Hokkaido (S. Imai). Type in Herb. Imai.

5. *Elaphomyces nikkoensis* Imai, in Proc. Imp. Acad. Tokyo **14** : 18 fig. A 1-4 (1938).

Crust almost none. Fructification 17×15 mm, black, densely distributed with papillate warts, which are partly irregular in arrangement. Cortex black, carbonaceous, fragile, 0.3-0.5 mm, difficult to make section of it. Peridium when matured forming very thin and white membrane inside of cortex. Ascospores sphaerical, black or dark brown, $23-30 \mu$ in diameter, with irregular and granulous surface formed of fine fasciculate spines.

Nom. Jap. Nikko-kurotutidango

Hab. Found without being parasitized by *Cordyceps*. Near summit of Mt. Ryogami, Prov. Titibu (Oct. 16, 1951, D. Shimizu) Among *Thujopsis dolabrata* forest, Nikko and Azumimura, Nagano Pref. (by S. Imai).

Only two fructifications were found without parasitic *Cordyceps*, in too much matured condition to research details of cortex and gleba. Original description by



Fig. 4. *El. nikkoensis*
Ascospore

S. Imai is as follows.

"Mycelio atro-brunneo, non copioso, facile separabili, terreno; ascomate globoso dein irregulariter globoso, 1-2 cm in diam., recente atro, sicco leviter fusciscente, cortice carbonaceo rigidoque, 0.5 mm. crasso, verruculoso, verrucis minute granulosis otusis; peridio recente 1.5-2 mm. crasso, sicco 0.5-1 mm. crasso, extus albidulo intus griseo, sicco grisescente, homoganeo; gleba initio albo-filamentosa, dein olivaceo-brunnea, demum nigrescente; venis sterilibus griseolis, evanescentibus; capillitiis hyalinis, tenuis 2-2.5 μ crassis; sporis globosis, atrobrunneis, opacis 22.5-32.5 μ in diam. verruculosis."

6. *Elaphomyces titibuensis* Y. Kobayasi sp. nov.

Crusta destituta. Fructificationes 15 \times 14 mm. Cortex nigrtellus dense verrucosus, areolatus, areolis polygonalibus 0.2-0.6 mm in diam. Verrucae minutae obtusae fere non costatae, in sectione conicae 200-240 μ alta, 230-260 μ lata, centraliter flavescens, prosenchymaticae, periphariter aurantiacae pseudoparenchymaticae. Cellulae tangentiales hyalinae 3-3.6 μ crassae. Textura apicalis verrucae nigrigella e cellulis radiatim dispositis elongatis obscure olivaceis composita. Peridium in sectione 1.5-1.8 mm crassum pallidum, inferne obscure olivaceum, non maculatum, dissepimentis rubescens, angustatis bene differentialis a peridio. Gleba obscure porphyreo-fusca, non cavitata.

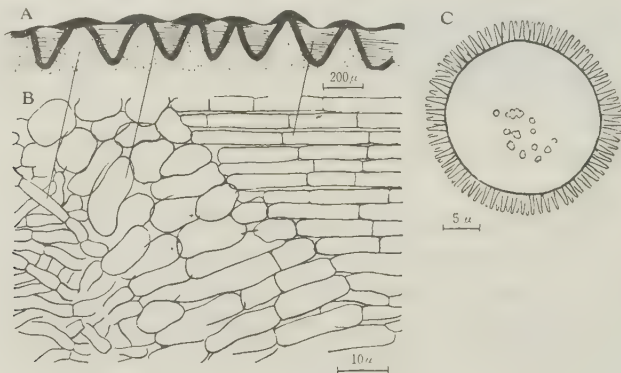


Fig. 5. *EL. titibuensis*

A, B. Section of cortex C. Ascospores

Ascosporae sphaeroideae 25-30 μ in diam., membranaceae vel nigrescentes, densissime spinosae, spinulis plus minusve coalescentibus. Capillitia hyalina, ramosa 3-5 μ crassa.

Mycelial crust none, mycelial strands scantily covering the surface of fructifications, which are 15 \times 14 mm in size. Cortex almost black, densely beset with low warts, showing polygonal areolae (0.2-0.6 mm in diameter). Warts small, not pointed, almost without ridges, in section, conic, 200-240 μ high, 230-260 μ broad, central part of slender, interwoven hyphae, pale yellow, outer part of layer pseudoparenchymatous

cells or thick hyphae, bright orange colour, tissue among warts composed of tangential cells of $3-3.6\ \mu$ broad hyaline hyphae, superficial tissue radiating from top of almost all warts less regularly arranged, dark olivaceous. Peridium in section $1.5-1.8\ \text{mm}$ thick, pallid, densely composed of hyaline $3.5-5\ \mu$ broad hyphae, inner part dark olivaceous, not marbled, dissepiments reddish, distinctly separated from peridium, narrow. Gleba dark purplish brown, stuffed. Asci unknown. Ascospores $25-30\ \mu$ in total diameter, dark brown or almost black, densely covered with conspicuous coalescent spines. Capillitia of $3.5\ \mu$ broad, hyaline branched hyphae.

Hab. Host of *Cordyceps ophioglossoides*. Mt. Nagasawa, Prov. Titibu (Nov. 10, 1951, D. Shimizu). Type in Herb. Nat. Sci. Mus.

In appearance, this resembles to *El. miyabeanus*, differing, however, in larger ascospores.

7. *Elaphomyces reticulatus* Vitt., Monographia Lycoperdineorum p. 107 pl. 3 fig. 10 (1842) et in Mem. R. Accad. Sci. Torino, ser. 2, 5 : 218 (1843); Fischer, in Rabenh. Krypt. Fl. Deutschl. ed. 2, 1 (5) : 93 (1897); Dodge, in Ann. Myc. 27 (3, 4) : 172 (1929); Lange, in Dansk Bot. Arkiv, 16 (1) : 23 (1956).

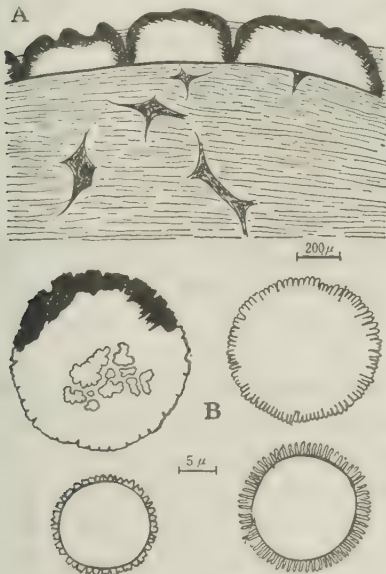


Fig. 6. *El. reticulatus*

A. Section of cortex and maculated peridium B. Ascospores

Fructifications almost globose, not depressed, medium-sized, $10-15\ \text{mm}$ in diam. $10-17\ \text{mm}$ high, partly covered with crust. Crust scanty, earthy with $0.2-0.3\ \text{mm}$ thick mycelial strang. Cortex ochraceous, warty; warts low, flattened, not pointed polyhedral with 3, 4, 5 ridges, at base covered with dark brownish tissue of tangential cells, in section with flat top, $200-250\ \mu$ high, $400-600\ \mu$ broad, yellow, pseudoparenchymatic, or partly prosenchymatic. Tangential cells white or pale yellowish, $3.7-4.4\ \mu$ thick. Peridium $2-3\ \text{mm}$ thick, pale vinaceous, distinctly marbled with reticulation of pale ochraceous veins, composed of $1.7-2.5\ \mu$ thick hyphae. Dissepiments pale ochraceous, distinct. Gleba black. Asci 4 spored.

Ascospores sphaerical $14-27\ \mu$ in total diameter, distinctly with thick, somewhat truncate spines, dark brown, showing irregularly verrucose appearance in adult stage. Capillitium not distinct.

Hab. Host of *Cordyceps canadensis*. In deciduous woods, Owizawa Pass, Owizawa, Yamagata Pref. (Sept. 16, 1958, Y. Kobayasi & D. Shimizu), Yanabazawa, Owizawa (Sept. 13, 1958). Nikko Bot. Gard., Nikko (Sept. 26, 1940, Y. Kobayasi).

Distr. Europe, U.S.A.

As already pointed out by Lange, this species can be distinguished by the flattened warts, paler peridium and the destitution of crust. It is interesting that Japanese fungi are all found as the host of *C. canadensis*.

8. ***Elaphomyces variegatus*** Vitt., Monographia, Tuberac. p. 68 pl. 4 fig. 4 (1831); Hesse, Hypog. Deutschl. **2** : 72 pl. 13 fig. 8-16 (1894); Dodge, in Ann. Myc. **27** (3, 4) : 167 (1929); Imai in Proc. Imp. Acad. Tokyo **15** : 146 (1939); Lange, in Dansk Bot. Arkiv **16** (1) : 22 (1956).

Syn. *Elaphomyces subvariegatus* Imai, in Proc. Imp. Acad. Tokyo **10** : 678 fig. 3-6 (1934).

"Fructifications globose or depressed globose, not rugose, 15-30 mm in diameter, ochraceous or becoming fulvous when dried. Crust earthy, composed of dark purplish mycelia, easily detachable. Cortex subcitrineous, not so differentiated from peridium. Warts small, 350-600 μ in diameter at base, 300-500 μ high, with 3-6 ridges, pointed or slowly obtuse. Peridium about 1 mm thick, reddish brown, variegated with brown reticulations like labyrinth. Gleba filamentose, white at first, then smoky avellaneous, at last becoming blackish brown and furfuraceous. Asci 4-8 spored. Spores spherical, blackish brown, 12.5-22.5 μ in diameter, echinate, epispore 1.5-2.5 μ thick."

Nom. Jap. Amine-tutidango

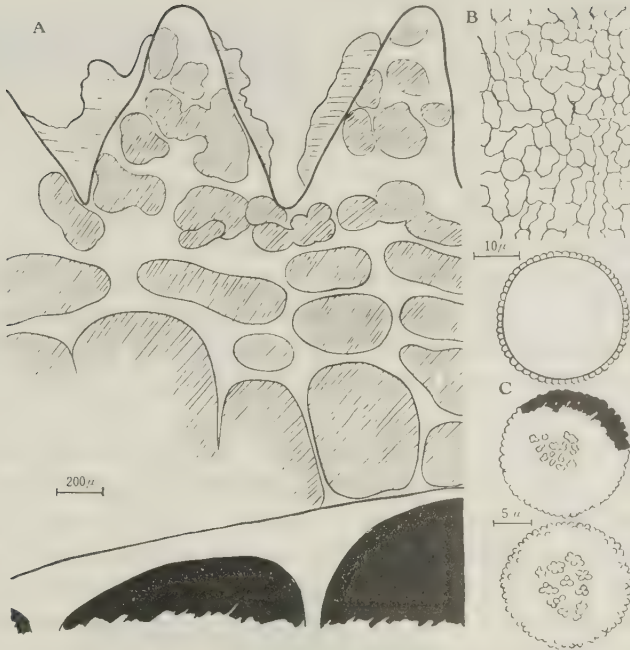
Hab. Host of *Cordyceps ophioglossoides* and *C. intermedia*. In woods. Prov. Ugo, Kôzukey, Sinano, Tamba, Suô, Honsyû. Mt. Kurodake, Prov. Isikari, Hokkaido (as *El. subvariegatus*).

Distr. Europe (incl. Moscow), North America.

This seems to be commonly found in temperate region of the Northern Hemisphere. Although Imai separated *El. subvariegatus* from *El. variegatus* by the smaller warts on the surface of fructifications and the thinner peridium which is less than 2 mm in thickness, the writer considers them conspecific. As the writer does not yet collect this species, the above description was quoted from that of *El. subvariegatus* by Imai.

9. ***Elaphomyces muricatus*** Fr., Syst. Myc. **3** : 59 (1829); Dodge, in Ann. Mycol. **27** (3, 4) : 169 (1929); Lange, in Dansk Bot. Arkiv, **16** (1) : 21 (1956).

Fructifications depressed globose, 23×19 mm. Crust none, mycelial strang scanty, 0.3-0.5 mm thick, brown. Cortex yellowish brown or fuliginous, very conspicuously warty, area among warts deep, almost black. Warts large pointed, brown or deep ochraceous with several ridges and polygonal base, in section 900-1,000 μ high, 800-900 μ broad, yellow or orange yellow, somewhat prosenchymatous, of 2.5-3.5 μ thick

Fig. 7. *El. muricatus*

A. Section of cortex, peridium and a part of gleba
 B. Tissue of cortex C. Ascospores

cells. Tissue of tangentially arranged 3-4 μ thick cells among warts scanty and partly none. Peridium 2 mm thick, externally continuous with cortex, internally defined definitely from gleba, dark vinaceous brown (Hay's Brown), marbled with whitish veins, paler outward, hyphae of outer layer hyaline, 2.5-3.5 μ thick, somewhat thick-walled with narrow lumen, hyphae of inner layer 2.5-3 μ thick, vinaceous and thin-walled. Dissepiments narrow, pale orange. Gleba dark purplish brown (Seal Brown) with large cavity. Asci uncertain. Ascospores sphaerical, 12-20 μ in diameter, dark purplish brown, densely papillate, papillae somewhat conical in adult stage. Capillitia slender 2-2.5 μ , hyaline.

Hab. Host of *Cordyceps ophioglossoides*. Mt. Wanagura, Prov. Titibu (Sept. 23, 1951, No. 337, D. Shimizu).

Distr. Europe, U.S.A.

Although many European authors have been described this species, these literatures can not be quoted without reexamining the authentic materials. There are some differences between the descriptions by C.W. Dodge and M. Lange. According to Dodge, the spines of fructifications are sharp and slender, ascospores 19-25 μ in dia-

meter, slightly rough, and Lange describes that spines are large, deep brown, ascospores $18-25\ \mu$, densely echinate. Japanese fungus seems to be the same with the description by Dodge in the marbled peridium and the ascospores, except for the colour of cortex which seems to be darker than that of European and American fungi. According to Dodge, the species with marbled peridium such as *El. muricatus* are enumerated as follows.

El. variegatus Vitt., with small, pointed warts.

El. reticulatus Vitt. with flat warts.

El. verrucosus Dodge with minute warts ($80-90\ \mu$ high).

El. decipiens Vitt. with small rounded warts, spores radially striate.

10. ***Elaphomyces fragilisporus*** Imai, in Proc. Imp. Acad. Tokyo **15** : 146 (1939).

"Crust abundant, black-brown, composed of mycelial strangs and earth, easily detachable from fructifications. Fructifications globose, subglobose or depressed subglobose, not wrinkled, $15-20\ \text{mm}$ in diameter, ochraceous, becoming fulvous when dried. Cortex $350-500\ \mu$ thick, carbonaceous, rigid, yellowish, verruculose, warts minutely granulous, low, rotundate, $0.2-0.3\ \text{mm}$ in diameter. Peridium about $1.5\ \text{mm}$ thick, more or less yellowish, homogeneous. Gleba when matured vinose-blackish, pulverate. Ascospores globose, brown, $30-40\ \mu$ in diameter, episporos very thin, easily ruptured, reticulated or verruculose. Capillitia hyaline, abundant, filiformed, about $2.5\ \mu$ thick."

Nom. Jap. Taisetu-tutidango

Hab. In woods, Isikari, Hokkaido. Host of *Cordyceps capitata* (*C. canadensis*?).

Known only from type locality. Type preserved in Herb. Imai.

According to Imai, the present fungus is somewhat related to *El. asperulus* Vitt., from which it is easily distinguished by the thinner wall of spores.

Granulatus-Group

This has many common characteristics as follows.

Cortex verrucose, commonly with low warts, yellowish, clay colour, ochraceous or cinnamon, never black or brown. Cut surface of peridium homogeneous, never marbled. Ascospores spiny, free or coalescent.

In this group are enumerated *El. granulatus*, *El. japonicus*, *El. shimizuensis*, *El. neoasperulus* and *El. asperulus*, last of which is not yet found in Japan.

11. ***Elaphomyces japonicus*** Lloyd, Mycological Notes no. 44 : 609 fig. 860 (1916); Imai, in Trans. Sapporo Nat. Hist. Soc. **11** : 34 (1929).

Crust distinct and thick, of mycelial strang and earth, strang $0.3-0.7\ \text{mm}$ thick, reddish brown, anastomosed, detachable from cortex. Fructifications depressed globose, $17-35 \times 11-25\ \text{mm}$. Cortex $0.5-0.7\ \text{mm}$ thick, hard, yellowish brown, clad with angular or irregularly rounded obtuse warts which are partly covered with tangential cells.

Warts in section $230-360\ \mu$ broad, $270-360\ \mu$ high, dark reddish brown, composed

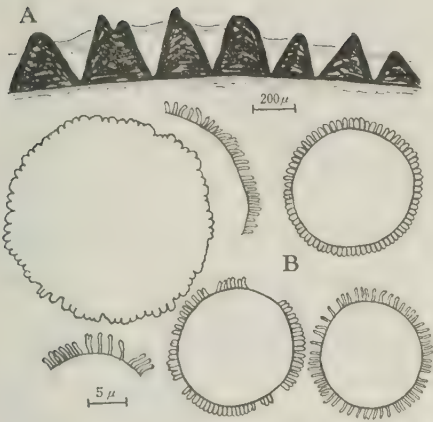


Fig. 8. Paratype specimen of *El. japonicus*
A. Section of wart B. Ascospores

Nom. Jap. Ki-tutidango

Hab. Host of *Cordyceps japonica*, *C. ophioglossoides* and *C. capitata* Okazaki, Aiti Pref. (J. Umemura-Type preserved in the Lloyd Herb. of the National Fungus Col-

of pseudoparenchymatous tissue. Tangential cells subhyaline, 2-3 μ thick. Peridium 2-3 mm thick, more or less soft, composed of whitish or pale yellowish outer layer, deep vinaceous brown middle layer and pale rosy inner layer. Dissepiments somewhat anastomosed into gleba, pale rosy. Gleba dark brown. Asci 5-7 spored. Ascospores 12-32 μ in diameter, commonly divided into two types; small type 12-20 μ , dark brown or almost black, large type 23-32 μ , brown, both covered with relatively low spines. Capillitia scanty, hyaline, simple or branched, 2-5 μ thick.

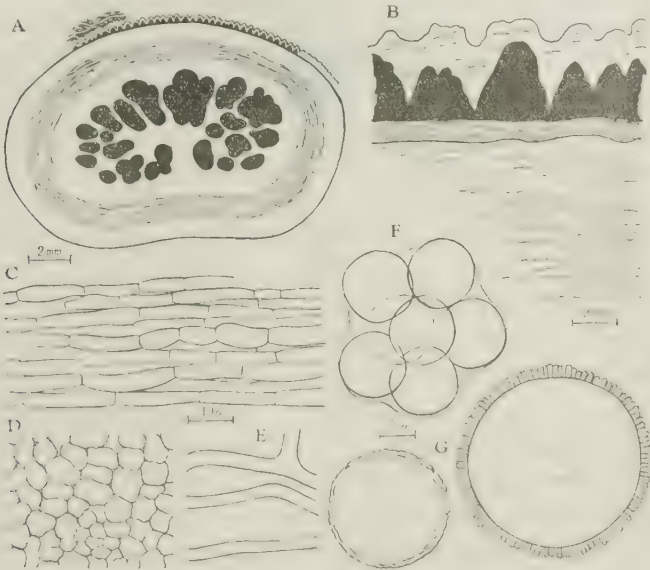


Fig. 9. *El. japonicus* (Specimen coll. in Oze)

- | | |
|------------------------------|-----------------------------------|
| A. Section of fructification | B. Section of cortex and peridium |
| C. Tissue of peridium | D. Tissue of wart |
| E. Capillitia | F. Asci |
| | G. Ascospores |

lection, U.S. Dept. Agriculture, Paratype preserved in Herb. Imai). Among *Tsuga diversifolia* forests, Ayamedaira, Oze, Gumma Pref. (Oct. 3, 1950, Y. Kobayasi).

According to the description by Imai, fructifications of *El. japonicus* have cinnamon colour, surface entirely covered with minutely poppy seed-like warts, cortex dull yellowish or light yellowish brown, separable from the peridium when dried; peridium light yellowish gray, pinky, about 0.5 mm thick when dried,..... spores $14-42.5\ \mu$ in diameter, finely echinulate.

In comparing with the paratype specimens preserved in Herb. Imai, the fungus collected in Oze has the thick peridium (2-3 mm) and larger warts. In spite of these differences, the writer considers these to be conspecific.

12. ***Elaphomyces granulatus*** Fr., Syst. Myc. **3** : 58 (1829), emend Hollós Magyarorszag Földalatti Gombai 76-77 : 198-200 (1911); Imai, in Proc. Imp. Acad. Tokyo **14** : 18 fig. B 1-4 (1938); Lange, in Dansk Bot. Arkiv **16** (1) : 18 fig. 4 a-c (1956).

Syn. *Elaphomyces cervinus* [L. ex S.F. Gray] Schlechtendal, Fl. Berol. **2** : 166 (1824); Dodge, in Ann. Myc. **27** (3,4) : 173 (1929).

Fructifications globose or depressed globose, 15-38 mm in diameter, 15-20 mm high, commonly covered with distinct earthy crust.

Cortex ochraceous, $350-400\ \mu$ thick, with low and somewhat pointed warts, easily detachable from peridium. Warts various in size, apically not so acute, with 3-4 ridges, polygonal at base, in section $180-250\ \mu$ high, $330-370\ \mu$ broad, internally brown, basally yellow, totally embedded in hyaline tangential cells. Peridium 1-2 mm thick, upper layer pale yellow, lower gradually becoming red brownish, not marbled. Dissepiments vinaceous, narrow at the upper side of gleba and broader at the bottom. Gleba dark purplish brown, pulverulent. Asci 5-7 spored. Ascospores in young stage covered with gelatinous sheath, densely spinose, spines very conspicuous $2-3.5\ \mu$ high, frequently fasciculate, total diameter $17-23-27\ \mu$, dark brown. Capillitia hyaline, somewhat thick-walled, $3-4\ \mu$ thick.

Nom. Jap. Tutidango

Hab. Host of *Cordyceps ophioglossoides* and *C. japonica*. Most commonly found among coniferous or deciduous forests in Japan. Ôdake, Mt. Hakkoda (Aug. 1953, Y.

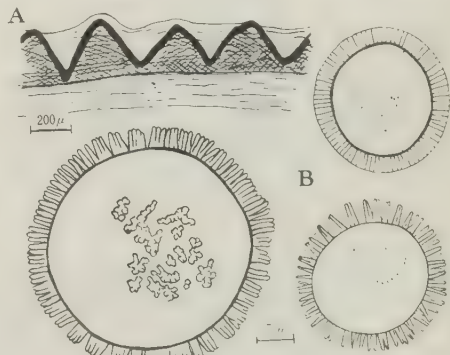


Fig. 10. *El. granulatus*

A. Section of cortex and peridium
B. Ascospores

Kobayasi); Isigakimati, Minamihara, Yonezawa (Aug. 27. and Sept. 21, 1958, D. Shimizu); Sirahata pine forests, Yonezawa (Sept. 20, and Nov. 30, 1958, D. Shimizu); Mt. Wanagura, Prov. Titibu. (Sept. 1950, D. Shimizu); Nikko (Imai).

Distr. Europe, N. America, probably cosmopolitan in the Northern Hemisphere.

13. *Elaphomyces shimizuensis* Y. Kobayasi sp. nov.

Fructificationes 11-22×10-18 mm, partim e crusta contegentes vel fere nudaе. Cortex ochraceus minute granulatus. Verrucae conicae 400-500 μ altae, 300-450 μ latae, apice acutae, flavae e cellulis tangentialibus contegentes. Peridium 1-1.5 mm crassum, por-

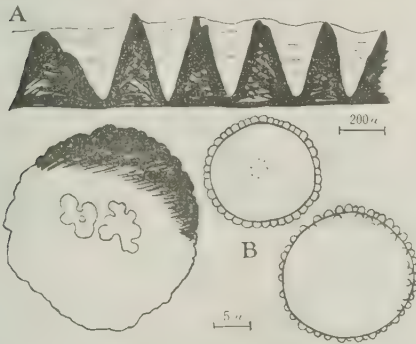


Fig. 11. *El. shimizuensis*

A. Section of wart B. Ascospores

phyreo-fuscum, non maculatum. Gleba bene differentia a peridio, dissepimentis angustis rufescentibus. Ascosporae 15-17 μ vel 25-27 μ in diam., nigrobrunneae, minute papillatae.

Crust poorly developed or almost none. Fructifications subglobose 11-22×10-18 mm. Cortex ochraceous, minutely granulate, fragile, warts almost embedded in tangential cells and hardly discernible by naked eye. Warts in section high conic, somewhat acute,

400-500 μ high, 300-450 μ broad, bright orange. Tangential cells pale yellow, 5-6 μ thick. Peridium 1-1.5 mm thick, not marbled, homogeneously vinaceous brown, composed of 3-4 μ thick hyphae. Gleba distinctly separated from peridium, with narrow and red coloured dissepiments, when matured forming cavity in the central core. Asci 7 spored. Ascospores divided in two different types, small type 15-17 μ , large type 25-27 μ in diameter, dark brown, almost black, minutely papillose, in adult stage irregularly verrucose.

Hab. Host of *Cordyceps ophioglossoides*. Mt. Wanagura, Prov. Titibu (Sept. 22, 1951, D. Shimizu). Type (no. 336) preserved in Herb. Nat. Sci. Mus.

In Granulatus group, this species is distinct by the papillose ascospores.

14. *Elaphomyces neoasperulus* Y. Kobayasi sp. nov.

Fructificationes depresso-globosae 22-25×16-20 mm, partim e crustis rufescentibus contegentes. Cortex 0.7-0.8 mm crassus, dense verrucosus, verrucis cinnamomeis pyramidalis, acutis, basi polygonalibus, maturitate faciliter separabiles e peridio. Verrucae in sectione acutae 580-750 μ altae, 330-500 μ latae, e cellulis luteis vel fusco-aurantiis compositae, e cellulis tangentialibus hyalinis contegentes. Peridium 2-2.5 mm crassum, non maculatum, coriaceum, superne pallide ochraceum, inferne vinaceo-fuscum, dissepimentis angustis. Gleba nigrobrunneae vel fere nigra. Asci cum 4-7

sporis. Ascospores nigrobrunneae vel nigra, 17–27 μ in diam., minutissime spinulosae, spinulis brevibus plusminusve tessellatim vel irregulariter coalescentibus. Capillitia hyalina, 3–5 μ crassa.

Crust poorly developed, of reddish brown mycelial strand and earth. Fructifications somewhat depressed globose, 22–25 mm broad, 16–20 mm high. Cortex 0.7–0.8 mm thick, distinctly and densely set with pointed and pyramidal warts with polygonal base, reddish brown, at maturity easily peeled away from peridium, leaving foveolate and tawny coloured scar on surface of peridium. Warts commonly pointed, in section 580–750 μ high, 330–500 μ broad, of yellow or brownish orange-coloured cells, covered with hyaline tangential cells. Peridium 2–2.5 mm thick, not

marbled, soft, leathery, upper layer 1 mm thick, pale ochraceous, lower vinaceous brown with several narrow dissepiments. Gleba dark brown or almost black. Asci with 4–7 spores. Ascospores dark brownish or almost black, 17–27 μ in diameter, very densely set with low spines which is coalescent making surface of spore as divided up in irregular blocks. Capillitium scanty, 3–5 μ thick, hyaline.

Hab. Host of *Cordyceps ophioglossoides*. Among *Pinus densiflora* forests, Sirahata, Yonezawa, Yamagata Pref. (Oct. 29, 1957, D. Shimizu). Type (no. Th. 4) preserved in Herb. Nat. Sci. Mus.

This resembles to *El. asperulus*, especially in the surface view of ascospores, but the latter species differs in small and somewhat flattened warts and larger (28–38 μ) ascospores.

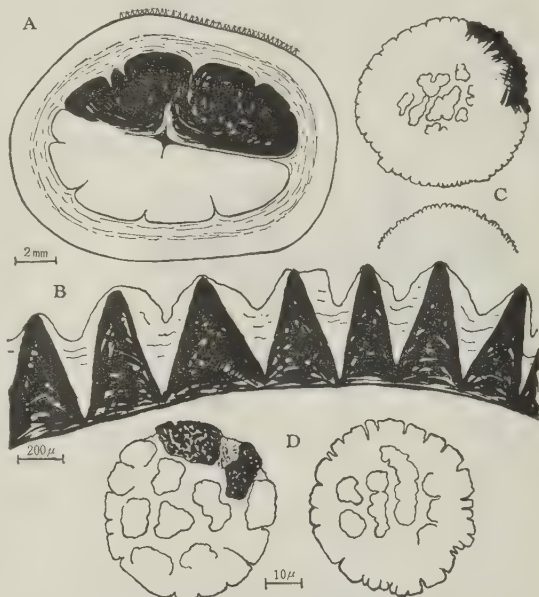
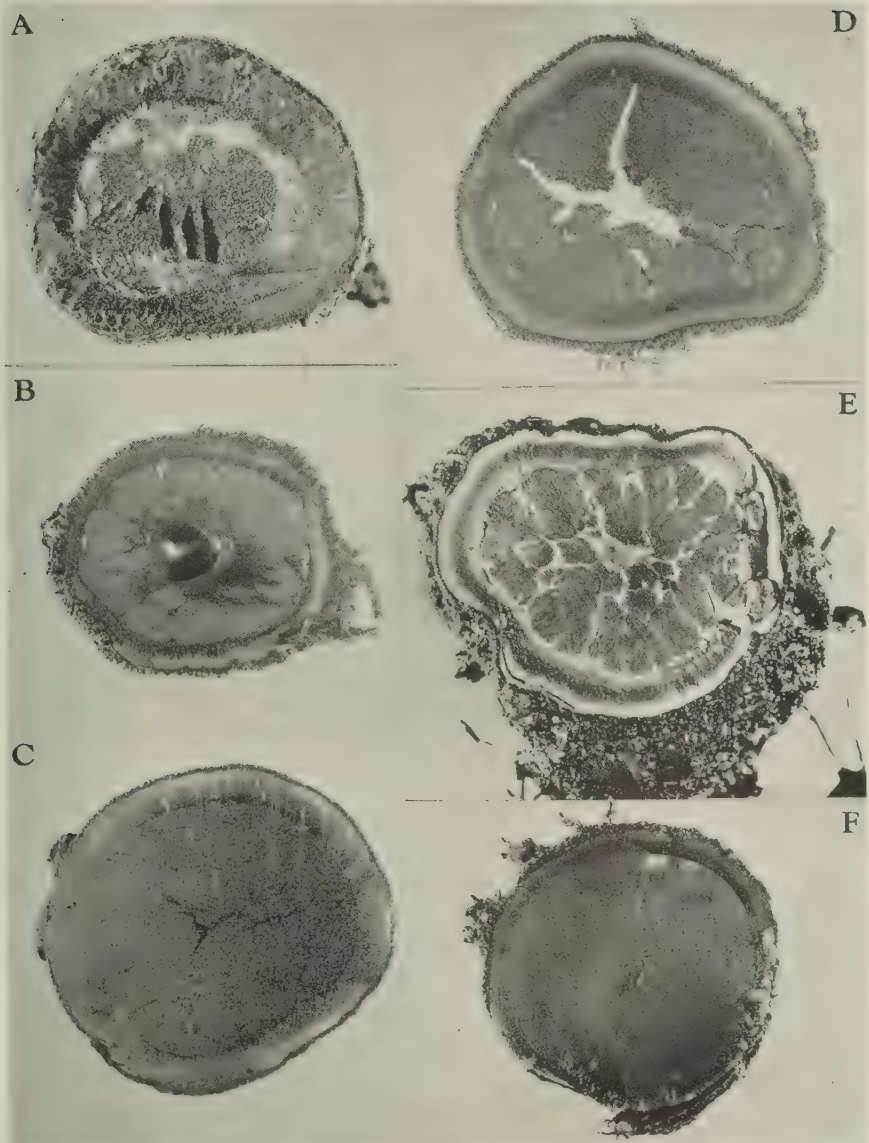


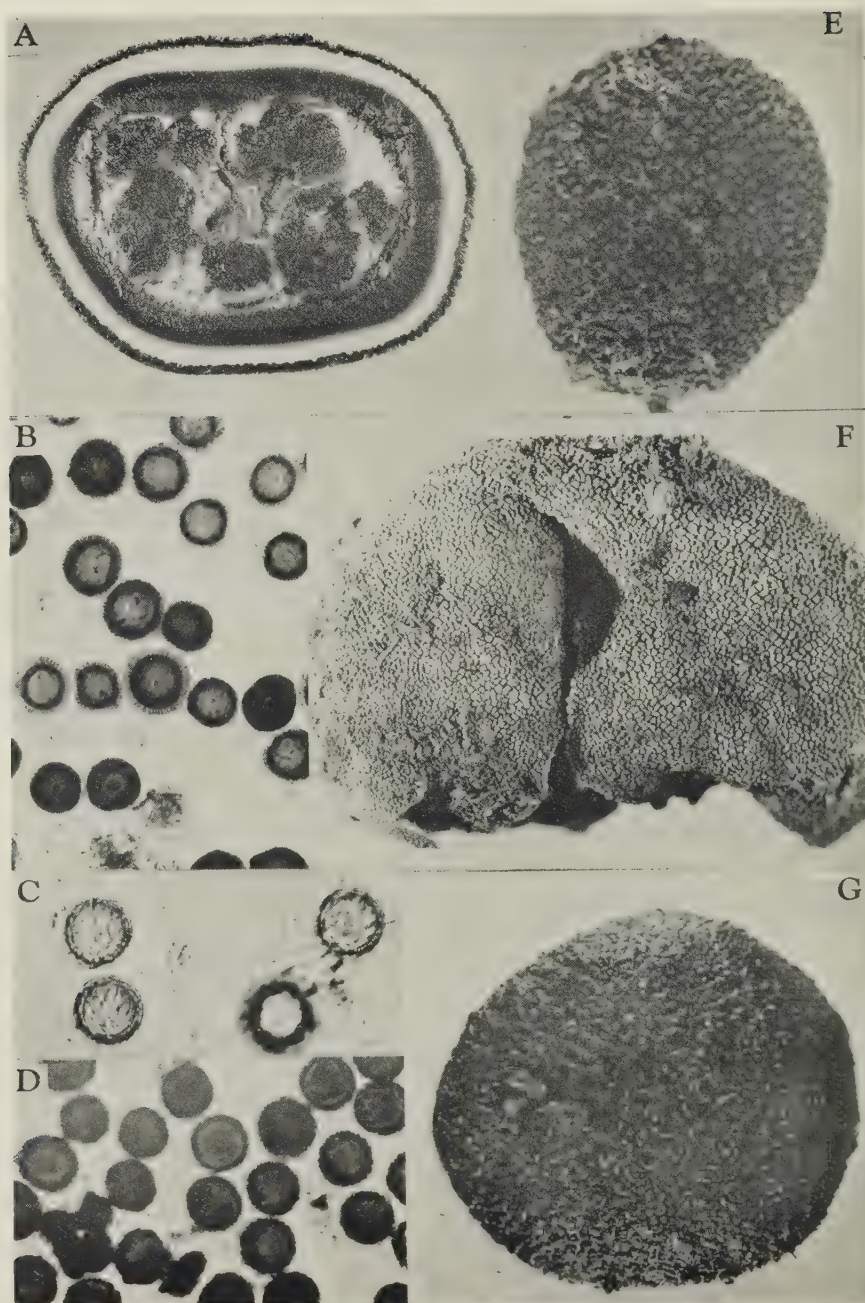
Fig. 12. *El. neoasperulus*

A. Section of fructification B. Section of wart
C. Ascospores



Pl. 5. Sections of fruitbodies of *Elaphomyces*

- A. *El. reticulatus* $\times 3.8$ B. *El. muricatus* $\times 0.8$
C. *El. titibuensis* $\times 2.8$ D. *El. neoasperulus* $\times 2.2$
E. *El. granulatus* $\times 2.5$ F. A form of *El. granulatus* $\times 1.7$



Pl. 6. A. Section of fruitbody of a form of *El. japonicus* (coll. in Oze) $\times 3.3$
 B-D. Ascospores B. *El. granulatus* $\times 450$ C. *El. asahimontanus* $\times 500$
 D. *El. muricatus* $\times 450$
 E-G. Surface view of cortex E. *El. muricatus* $\times 2$ F. *El. japonicus* $\times 3$
 G. *El. neoasperulus* $\times 2.5$

趾間部病巣より得た *Cephalosporium* について

On a species of *Cephalosporium* isolated from the epidermophytosis between the toes.

原 田 誠 一*, 臼 井 良 照**

Seiichi HARADA and Yoshiteru USUI

緒言：*Cephalosporim* は分生子柄が菌糸より直立し、その先端に分生子を順次発生するが、互に粘着して小球状をなす不完全菌を総称する属名である。病原菌として世界各地より約 20 株が培養されているが、我国では上林¹⁾ (1937)、森川¹⁾ (1939)、高橋(吉)、黒田²⁾ (1949)、百瀬、深道³⁾ (1955) の報告をみるのみで、稀有のものといえる。我々は最近臨床上趾間白癬の像を呈する病巣より本属の菌を得、純粋培養による形態学的検討を加えたのでここに報告し、多少の考察を加えようと思う。

症例：患者、小林某、34 才♂、会社員。初診、昭和 35 年 8 月 8 日。

家族歴、既往歴に特記事項はない。同胞 2 人健在である。

現症歴、約 3 年来右第 4 趾間部に所謂水むしを生じ、塗薬を塗布したが一進一退、最近は何と痒通が甚しくなったので来院した。

現症、右第 4 趾間は硬く肥厚し淡褐色、胼胝状を呈する。これを剝離すれば、その下に白色の浸軟せる状態をみるが、湿潤することはない。この所見は我々が通常みところの趾間白癬と特に異つたものではない。病巣は右第 4 趾間に限られ、その他の部位には全く変化がない。(Pl. 8-1)

即席検鏡：趾間の淡黒褐色の部を除き白色の浸軟部をとつて検鏡すると、やゝうねりの強い菌糸が見出された (Pl. 8-6)。芽胞と思われるものは見当らず、連鎖芽胞もみることはできなかった。

培養：病巣浸軟部の小片をサブラウ含糖培地に移植すると 3 日目に湿性のコロニーが発生した。発育は比較的速で、4 日目には 15×14 mm 大、白色の空中菌糸の発生がある。9 日目には綿の如き感を呈し、中心もり上りボタン状、深部は黄褐色、空中菌糸の発生が盛である。周辺部は湿性匍匐性菌糸をなし、裏面は美麗な橙色を呈する。約 2 週間後にはコロニー斜面全体を蔽い、淡灰色、中心菌糸束、つぎにやゝ褐色、周辺平坦、全体に占綿状をなす。その後日を迫るに従い空中菌糸の発生著明となりコロニー全体はこげ茶色を呈し、点々凝固水をのせる。裏面は赤褐色をなす。麦芽浸出液寒天上の発育は良好で、拡散性、始めは白色であるが次第に淡褐色を呈し、占くなると胞子形成部分は淡褐色の色調が強い。裏面は淡褐色を呈する。

サブラウ葡萄糖寒天における巨大培養 1 週間後の所見は、径約 7 cm 大、やゝ淡黄白色を呈し中心ボタン状となり菌糸束の発生があり、これより褐色の放射状溝が周辺に向つて同心円状に拡かる。裏面は中心星状の黒褐色の部があり一般に淡黄赤色を呈する。(Pl. 7-3)

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懸滴培養, サブラウ葡萄糖寒天における所見菌糸は隔壁を有し分岐する。集合して束状をなすところもある。分生子柄は一般に菌糸より直立しその先端に分生子をつける。時に分生子柄は直立しないものもあり, 時に分岐し分生子柄と菌糸との間にはその基底に隔壁を有し或はこれを欠く。分生子柄は 80μ 前後の長さを有し, 隔壁を有するものがある。又ラケット様菌糸が見られる。分生子は集団をなしその数, 十数個に達する。集団の径 48μ に及び, 或は分生子極めて少く僅かに $1 \sim 2$ 個のものも時としてみられる。分生子はやゝ孤状をなし単室又は2室に分れることがあるがそれ以上多房状を呈することはない。長径 $8 \sim 16 \mu$, 短径 4μ 。分生子が菌糸の側縁に集合する状態もみられ, この場合は石垣状に積み重なる。クラミドスポール (chlamydospore) は古き菌糸に類しくみられ $8 \times 16 \mu$, 単室又は $2 \sim 3$ 室に分れ猩紅色菌における大分生子の如き観を呈し, 2重の輪廓と顆粒を容れ光線を強く屈折する。菌糸, 分生子柄, 分生子とも着色することはない。又分生子の発生は先端より順に大となつて分生子柄に直角につく場合もある。(Pl.7-1, 2, 4)

麦芽浸出液寒天においては菌糸は巾 $2.5 \sim 3.0 \mu$ 滑面, 透明で隔壁があり不規則に分岐する。培養時間を経るに従い油滴を内蔵する。分生子柄は長さ不同 $30 \sim 150 \mu$, 巾 2μ , 滑面, 稀に分岐して隔壁を生ずることがある。先端から分生子を生じ粘質物により約 30μ 径の塊状体として着生する。分生子は楕円形又は腎臓形で曲折したものが多い。滑面透明, $9 \sim 15 \times 3.5 \sim 5 \mu$, 古くなると分生子のなかには2細胞となるものがあり $13.7 \sim 25 \mu$ 又はそれ以上に達する。クラミドスポールは菌糸端に生じ, 厚い膜をもち単又は2細胞, 顆粒に富み幾分黄色調を有し巾 10μ , 長さ 15μ 内外である。

考按: 本例は培養の所見よりすれば *Cephalosporium* Corda に属すべきである。一般に *Cephalosporium* によつて惹起される病変は Cephalosporiosis と呼ばれ世界各地から報告されている。本邦における症例は上林 (1937) の *C. nigrum* Kambayashi があるが黒色菌であつた。森川 (1939) は足の疣状皮膚炎及び趾間白癬を兼ねた1例から本属の菌を得 *C. keratoplasticum* Morikawa とし分生子は卵形又は楕円形。*C. arcuatum* Takahashi (1949) は高橋 (吉)¹⁾ によつて爪真菌症から又同じ菌株は高橋, 黒田²⁾ によつて趾間白癬様病巣から得られている。高橋はその菌株の分生子がすべて孤状をなすことより新種とし上述の如き名称を与えている。百瀬, 深道³⁾ (1955) は角膜真菌症から本属の菌を得て *Cephalosporium* sp. (Momose, Fukudo: 1955) として種名を決定せず報告しているが高橋によれば *C. arcuatum* と同一範囲に属すべきものであらうとしている。我々の分離した菌は分生子がすべて孤状を呈するというものではないので *C. arcuatum* と同一とは考えられない。分生子の形状, 大きさ等からして *C. keratoplasticum* Morikawa⁴⁾ に含まれるものと見做すのが妥当であらう。本菌は未だ精密な純粋培養によつて形態的研究が無く, 本報告をもつて嚆矢する。本属の菌は白癬様病変を起すことが多く, 又足菌腫を発生した報告がある。足菌腫から分離されたものは *C. recifei* Leao et Lobo (1934), *C. faliforme* Carrion (1951), *C. granulomatis* Weidman et Kligman (1945) 等がある。こゝで趾間部の病変であるが, 一般に趾間部からは種々な菌が検出される。即ち最も多いものは白癬菌で, つぎにカンデダである。趾間部の病変から全く糸状菌の検出されない場合も予想外に多い。しかも臨床所見は甚だしく相似していて, 単に臨床所見のみをもつて診断を下すこと

は不可能である。我々の例の如きもその一つであつて、外觀は趾間白癬に異ならない。趾間病変の正確なる診断は単に直接検鏡のみならず、培養をも遂行すべきであることを強調したい。

本菌の同定に當つては東北大学高橋吉定教授、長尾研究所椿啓介博士の御教示を得た。こゝに記して謝意を表する。

SUMMARY

A fungus of *Cephalosporium* isolated from the epidermophytosis between the toes was investigated morphologically under pure cultur.

The conidia of our fungus are oval, ellipsoid or sickle-shaped, uni-celled, often two-celled, characteristically and develop apically on the conidiophores which are straight mostly, rarely branched monopodially. In addition, chlamydospores develop in abundance terminally on hyphae when matured.

Because of the above reasons, the present fungus was considered to be classified as *Cephalosporium keratoplasticum* Morikawa, differing from *C. arcuatum* Takahashi, causing similar Cephalosporiosis, which forms sickle-shaped conidia.

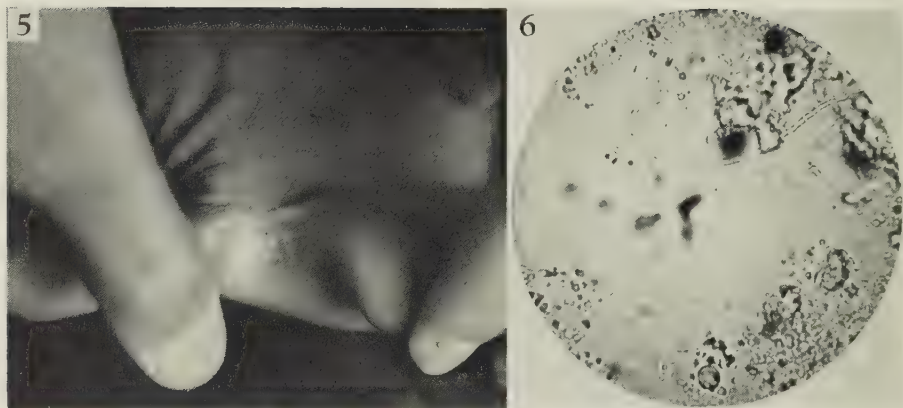
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Pl. 7. *Cephalosporium keratoplasticum* Morikawa

1. Conidiophores and uni-celled conidia (\times ca. 500)
2. Two-celled conidia (\times ca. 570)
3. Colony on Sabouraud-agar plate, after 10 days, at 30°C ($\times 0.6$)
4. Chlamydoconidia (\times ca. 500)



Pl. 8. *Cephalosporium keratoplasicum* Morikawa
5. Epidermophytosis between the toes ($\times 0.6$)
6. Microscopical figure of the region.

研究所員業績(1959-1960)

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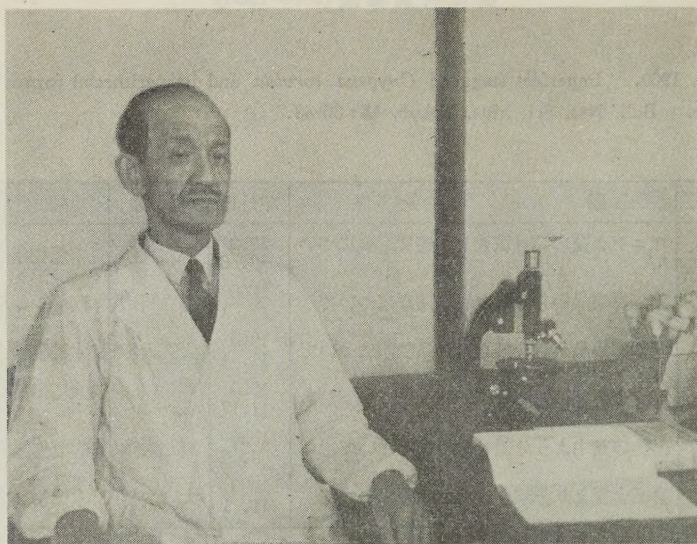
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“	南極土壌より分離された酵母菌について	1960 11. 1	大阪大学	日本植物学会25回大会

研究状況

研究員	題 目
渡辺 俊雄	各種線虫及びその他害虫に対する防除剤に関する研究
平邑 延康	
鈴木 義之	<i>Curvularia</i> 属菌の代謝産物に関する研究
“	大豆蛋白質に関する研究



† Dr. Kendô Saito, 1878-1960

長尾研御在職当時の故斎藤賢道博士(昭17)

昭和 35 年 10 月 15 日当研究所顧問、大阪大学名誉教授、理学博士斎藤賢道先生は忽然として逝去せられました。まことに得難き碩学を失い、痛惜の念に堪えません。謹しんで哀悼の意を表します。

我国建国 2600 年祝典挙行政に際し、その記念事業として応用菌学研究機関及び菌株保存施設を目的として、長尾研究所が設立せられんとするや、先生はこの企画に対し全面的に賛意を表せられ、諸般の協力を惜しまず、先生が多年に亘り蒐集せられました貴重なる菌株標本を中核に一千余種に及ぶ研究所保存菌株を整うことを得て研究所開設の運びに至りましたのは昭和 15 年 11 月 1 日でありました。爾後、先生は財団の理事として、又研究所の主任研究員として昭和 24 年まで在任せられ、其の後は顧問として今日に至られたのであります。

来る昭和 36 年 1 月には本研究所が財団法人の認可を得ましてから、満 20 週年に相当しますので、先生にもその祝賀の喜びを共にして戴きたいと一同楽しみにしていたのでありますが、俄かに幽明境を異にせられ、今は其の意を果たすべき術も絶えてしまいました。惜しみても余りある悲しいことであります。

先生が遺されましたあの菌株の蒐集は、現在もなお、長尾研究所に於て、植えつがれ、植えつがれて生き続けており、若き研究員たちはこれを長尾研究所の誇りとも財宝ともして護り、また、自然の本質の深奥を示唆する貴重なる研究対象としてこれと真剣に取り組んでおるのであります。結晶が核を中心として発育して行きます様に、長尾研究所保存菌株は先生の御蒐集を中核に逐年其の数を増し、また国際的の接触を拡げつゝあります。

先生の面影は、この菌株とともに、長尾研究所のあらん限り、研究員たちの心の中に生き続けて行かれることでありましょう。

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